THE PREVALENCE OF ANTIBIOTIC-RESISTANT BACTERIA IN STREET-VENDED FOODS

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DECLARATION OF INDEPENDENT WORK

I, Malerato Moloi, hereby declare that this research project submitted to the Central University of Technology, Free State for the degree MASTER OF HEALTH SCIENCE: ENVIRONMENTAL HEALTH is my own original work. This research work has not been submitted before to any institution by myself or any other person in fulfilment of requirements for the attainment of any degree or qualification.

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MALERATO MOLOI **DATE**

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SUMMARY

In recent years, the street-food vending industry has expanded globally, especially in developing countries and the expansion continues (Bereda *et al*., 2016). This industry has played a significant role in the socioeconomic development of these countries. In addition, it has a positive impact on urban dwellers as the majority of them depend on these foods for convenience (Bereda *et al*., 2016). Despite the economic benefits of street foods, there is a need to reflect on the hazards imposed by foods prepared within the food-vending industry. These foods may play a role in the transmission of disease-causineg microbes within a population (Ogidi *et al*., 2016) that may exhibit antibiotic resistant profiles.

Several studies have been carried out in South Africa to establish the relationship between food handling and the occurrence of food contaminants. However, few studies have been conducted in the Free State, therefore this area formed the basis of this study. This study had three aims, the first of which was to assess hygienic practices of the street food vendors and secondly, to evaluate factors that contributed to contamination and isolated microbes from surfaces and foods. This was followed by an assessment of the susceptibility of isolated microbes towards antibiotics. The study was carried out in Mangaung Metropolitan Municipality.

In the current study, the data on the hygiene practices and potential risk factors was collected using questionnaires and an observation checklist. The results obtained indicated that even though the vendors had a positive attitude towards food safety, non-compliance with food safety regulations was observed. Some of the vendors did not wash their hands and did not wear aprons during the processing and serving of food. Moreover, the vendors reported that they had not received any training in food safety and hygiene. It was also observed that the material used for the construction of the stalls was not able to protect the food from dust, given that all the stalls were situated by the roadsides, which were dusty and prone to fumes from motor vehicles. Furthermore, a lack of sanitation facilities was also a major problem identified.

Total viable counts were recorded on food preparation surfaces at various vending stalls in Thaba Nchu, Bloemfontein and Botshabelo. Microbial counts obtained in Botshabelo ($1.1x10^4$ to $1.1\ x10^6$ CFU/m^2) showed higher microbial counts as compared to those in Bloemfontein ($1.1x10^4$ to $1.1\ x10^5$ CFU/m^2) and Thaba Nchu ($1.1x10^4$ to $1.1\ x10^5$ CFU/m^2). These counts were found to be higher than the national standard (100 CFU/m^2). Other than food preparation surfaces, microbial counts of meat samples obtained in Bloemfontein, Thaba Nchu and Botshabelo were also found to be lower than the national standard (100 CFU/g). Nevertheless, the results showed the presence of *S. aureus* and *E. coli* indicates poor hygiene. This indicates that improper food handling practices were somehow carried out by food handlers, thereby contributing to the presence of these foodborne pathogens.

The predominant species identified were *S. aureus* and *E. coli*. The presence of these species is a major cause of concern because *S. aureus* and *E. coli* are the most common bacteria that play a significant role in human diseases (Setlhare, 2013).

In addition, when antimicrobial susceptibility testing was performed, *S. aureus* isolates showed resistance to gentamycin (100%), streptomycin (63%) and tetracycline (68%) which would make the treatment of infections with this species difficult. Moreover, 62% of *Shigella spp.* showed resistance to ampicillin, followed by *Escherichia hermannii* which showed 50% resistance to ampicillin. Lastly, *E. coli* showed 100% resistance to ampicillin. This indicates that ampicillin will no longer be useful for treatment of any infection caused by *Shigella, E. hermannii and E. coli*. As these bacteria have developed resistance against the tested antibiotics, food contamination with antibiotic-resistant bacteria can also be a major threat to public health, because antibiotic resistance determinants can be transferred to other pathogenic bacteria potentially compromising the treatment of several bacterial infections (Eromo *et al*., 2016) within a population. Results from this study also indicate that the general public can easily be exposed to antibiotic-resistant bacteria daily through conventional food intake because the street vendors are non-complaint with food safety hygiene practices. Exposure of consumers to antibiotic-resistant bacteria may make them vulnerable to various food diseases caused by antibiotic-resistant pathogens.

CHAPTER 1

PREVALENCE OF ANTIBIOTIC-RESISTANT BACTERIA IN STREET-VENDED FOOD

A LITERATURE REVIEW

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1.1 General background

Von Holy and Makhoane (2006) describe street-vended foods as "…foods and beverages prepared and/or sold by vendors in streets and other public places for immediate consumption or consumption at a later time without further processing or preparation". The foods are usually sold in clusters around places of work, schools, hospitals, universities, railway stations, bus terminals and taxi ranks in the urban areas (Von Holy *et al.*, 2006). The street-food industry plays an important role in developing countries in meeting the food demands of the urban dwellers (Bereda *et al*., 2016) and the economic needs of those selling. This type of food feeds millions of people daily with a wide variety of products that are relatively cheap and easily accessible (Bereda *et al*., 2016). However, foodborne illnesses of microbial origin are a major health threat associated with street foods (Bereda *et al*., 2016). Food contamination with antibiotic-resistant bacteria can also be a major threat to public health, since the antibiotic resistance determinants can be transferred to other pathogenic bacteria potentially compromising the treatment of severe bacterial infections (Eromo *et al*., 2016). Many of the bacterial species (such as *Salmonella, Campylobacter* and *Escherichia coli*) carried by animals can also cause disease in people (World Health Organization, 2017). These bacteria, which are frequently antimicrobial-resistant, can contaminate the food supply from farm to fork during slaughter and processing (WHO, 2017). If these bacteria become resistant to antibiotics, it will become difficult to treat them and more people could die from foodborne diseases (WHO, 2017).

According to a guide to producing safe food of animal origin for the South African cosumer (Undated), food can become microbiologically hazardous to the consumer when the principles of hygiene and sanitation are not met. Additionally, it becomes contaminated by pathogens from humans or from the environment during production, processing or preparation . It also becomes contaminated when it originates from a sick animal, for example, a cow with mastitis or an animal with anthrax . Contaminated food may become a vehicle for the transmission of diseases such as salmonellosis or staphylococcal food poisoning.

To prevent street food contamination, the Regulations Governing General Hygiene Requirements for Food Premises and the Transport of Food (Department of Health, 2002) states that any person working on food premises must be adequately trained in food hygiene by an inspector or any suitable person. However, Siddabathuni (2019) reported that many/most of the street food vendors are not trained and lack knowledge regarding the transmission of foodborne infections. Food hygiene consists of several principles adopted to ensure food safety and to protect food from any chemical, microbiological or other type of contamination that can render it unfit for human consumption (Hilario, 2015; WHO, 2011). Additionally, food hygiene rules and regulations are meant to prevent the spread of communicable diseases associated with food and food processing and to ensure that consumers are not victim to food fraud (Hilario, 2015; WHO, 2011). Proper food hygiene should ensure that food is handled, stored, prepared, and served in such a way and under such conditions to prevent, as far as possible, the

contamination of the food (Hilario, 2015; WHO, 2011). Food safety hazards could be physical, microbiological, or chemical; however, microbiological hazards pose a bigger challenge to the safety of street food since invisible, potentially harmful microorganisms can grow rapidly in street food owing to lack of proper storage facilities and subsequently cause foodborne diseases in consumers (Akinbode *et al*., 2011). Food is described as a silent vehicle for microbial, chemical and physical hazards to the consumer (Lamin, 2017).

1.2 Contamination of street food

Food prepared in unhygienic conditions may result in contamination and possibly result in the presence of microbial contaminants (Kariuki, 2018). One of the major health hazards associated with street foods is microbial contamination, leading to foodborne illness (Kariuki, 2018). Factors involved in contamination of food by microbes include poor food handling and preparation practices, inadequate food storage methods, the personal hygiene of the food vendors, lack of suitable ways for disposing waste and poor sanitation facilities (Kariuki, 2018). The World Health Organization (WHO) (2016) stated that foodborne diseases remain a major public health concern globally. In the United States of America (USA), it is estimated that 48 million people become ill from foodborne diseases (Centers of Disease Control and Prevention, 2016). In Africa, it is estimated that 92 million fall ill and 137 000 die each year (WHO, 2015). Annually, thousands of people in South Africa suffer from foodborne illnesses, usually involving severe diarrhoea, vomiting and stomach cramps because of eating or drinking contaminated food (Gordon-Davis, 2010).

Several researchers (Mensah, 2002; Tambekar *et al*., 2008; Khairuzzaman *et al*., 2014) have identified the existing processing methods used in street food preparation such as inappropriate holding temperatures, and poor personal hygiene of food handlers as some of the main causes of microbial contamination in street foods. Therefore, if street foods become contaminated, the people who consume them are exposed to and risk contracting foodborne diseases such as salmonellosis, listeriosis, typhoid fever, cholera, and diarrhoea (Manguiat *et al*., 2013; Liu *et al*., 2014). The large number of urban consumers who rely heavily on street food, including the perishable nature of the products being vended and the fact that the foods are in a state that is ready for consumption, make adequate control of the street food trade imperative for protecting public health. The fact that the foods are 'ready-to-eat' (RTE) means that under normal circumstances the consumer does not subject the food to further treatments that could serve to reduce levels of contamination prior to consumption.

Some of the factors that contribute to contamination of street food are insufficient reheating and cooling of food items and a long period of time between when the food is prepared and when it is consumed (Kariuki, 2018). These vending practices and facilities used for dispensing street foods were recognised in the literature as major contributors to the cross-contamination of street foods. Bryan (1998) also reported that major contamination of street foods occurred at vending sites because of cross-contamination during cutting and chopping. Instances where the same knife was used to cut and chop different food on the same surface without cleaning in between were observed by

Mosupye and Von Holy (2000). Additionally, the vendors handled food with their bare hands and even exchanged knives with fellow vendors without in between cleaning, thereby increasing the chances of cross-contamination. Hands are reported as the most significant source to transfer microorganisms from faeces, face, skin, or other sites to vendors (Balkaran *et al*., 2014). In this instance, vendors can be carriers of foodborne pathogens such as *E. coli, Salmonella, Shigella, Campylobacter* and *S. aureus.* They eventually transfer these foodborne hazards to the consumers, posing a risk to public health (Rane 2011, Balkaran *et al*., 2014).

In a study conducted by Khuluse (2016) it was observed that food preparation and cooking space was minimal; as a result, the researcher observed that in some stalls white and red meat were grilled on the same griller which increases the chances of cross-contamination. Ideally a food stall must have adequate space for the activities being carried out, and the layout should be set up to ensure the segregation of non-ready-to-eat and ready-to-eat food (Food Safety Authority of Ireland, 2016). Additionally, Regulations Governing General Hygiene Requirements for Food Premises and the Transport of Food, GNR. 918 of (1999) state that the location and design of food premises shall be in a way that they will not create any health hazard. The risk of contamination during food preparation in external environments can be excessive owing to the lack of infrastructure and adequate resources (Sabuj *et al*., 2018). Additionally, waste and garbage produced in restaurants and discarded nearby can attract rodents and insects

that may harbour foodborne pathogens, as well as attracting flies that land on food sporadically (Sabuj *et al*., 2018).

The above-mentioned practices highlight risks that may exist in the street-vending business. Besides these practices, one of the biggest factors that influence street-food safety is provision of clean water at street-food stalls (Thanh, 2015). Studies carried out in different regions of Asia, Africa and South America have frequently highlighted the unavailability of potable water for various activities at the vending site as a major concern (Rane, 2011). Safe water is described as an essential requirement for health (Thanh, 2015). Owing to the shortage of clean potable water at vending sites, hand and dishwashing water is often reused, sometimes without soap (WHO, 1999). Proper hand and dish washing should be done with soap under running water (Mlay, 2018). In situations where dishing bowls are not washed regularly, an environment is created for insects such as flies to hover around to contaminate food (Mlay, 2018). Moreover, in most cases, street vendors store water under vulnerable conditions that can be subjected to contamination (Mjoka *et al.,* 2017). Without enough water, hygiene and sanitary practices cannot be exercised correctly.

Other real risk factors for contamination of food include the practice of under-heating to avoid shrinkage of meat and the initial contamination of raw materials with pathogenic bacteria or subsequent contamination by the vendors themselves during preparation (Oguttu, 2015). Several studies that investigated the cause of food poisoning indicated

that time and temperature abuse and cross-contamination were amongst the causes of food poisoning (Park *et al*., 2010; Baş *et al*., 2006). The growth potential of microbes is enhanced or increased through time, temperature abuse and cross-contamination (WHO, undated). Cross-contamination occurs when harmful micro-organisms are spread between food, surfaces, and equipment (Lamin, 2017). Additionally, the prevention of cross-contamination through the separation of raw and cooked foods during storage and preparation is an additional important consideration (Lamin, 2017). A long holding period of more than six hours, sometimes at ambient temperature, was reported to be a common factor contributing to foodborne illness through the multiplication of microorganisms favoured by a holding temperature in the range of 5 and 60 degrees Celsius (described as a danger zone) (Alimi, 2016). The Food Safety Authority of Ireland (2016) states that foods that are reheated at the stall must be heated to a core temperature of ≥ 70 degrees Celsius. This is done to avoid the risk of contamination in so far as is reasonably practicable.

In addition, other risks that contribute to the contamination of street food include the lack of proper utensils for cooking and the storage of prepared food. Poor quality of material coupled with improper practices may lead to the spread of toxins, pathogen growth or recontamination (Rane, 2011). The design, construction and maintenance of equipment and utensils are very important to food safety as their poor maintenance may lead to the inability to clean and sanitise surfaces effectively. This may then result in the build-up of residues of food, facilitating microbial growth and leading to an increased likelihood of

contamination. The appropriate use of equipment is also important to prevent the cross-contamination from raw materials (Rane, 2011).

1.3 Environmental hygiene status of street-food vendors work area

According to Kariuki (2018), vendors position themselves in overcrowded areas to reach out to large numbers of likely potential customers, hence leading to limited access to some of the basic sanitary facilities. Garbage and wastewater are typically discarded in the streets nearby, thus attracting rodents and insects and providing food for them which can serve as vectors for microbes. Street-food vendors can generate wastewater when washing hands, containers, and utensils and when cooking or preparing food. Water used for washing utensils and/or cooking or preparing food will contain fat, oil, and grease while water used for washing hands will contain soap and possibly particles and bacteria (Kariuki, 2018). It is argued that limited access to running water and waste disposal facilities increases the risk of foodborne disease (FAO and Department of Health, 2002).

Open bins are commonly used for garbage collection (Amlin, 2016). However, a study conducted by Hilario (2015) in Manila, Philippines reported that most (60%) vendors do not have garbage bins, hence they disposed their garbage near the stalls. Also, 74% of the vendors throw waste-water and solid waste next to the stalls, rendering the environment surrounding the eateries quite filthy. Studies have shown that vendors are not aware of a nedd to properly clean an area and segregate waste (Hilario, 2015). In

these areas grease is evident on the floor of the stalls owing to spills of oil and sauces (Hilario, 2015). If waste is not managed properly, it is not only aesthetically offensive, but it also poses risks to public health and clean-up can be costly. Moreover, litter from these sources easily ends up in sewers which can lead to the clogging of drainage systems (Cabaltica *et al*., 2016). According to Regulation (EC) No 852/2004 of the European Parliament and of the Council of 29 April 2004 on the hygiene of food stuffs, all waste is to be eliminated in a hygienic and environmentally friendly way in accordance with community legislation applicable to that effect and is not to constitute a direct or indirect source of contamination. Mangaung Local Municipality By-Laws Relating to Street Trading (2006) also stipulate that everyday street vendors must remove from any public road or public place at the conclusion of selling all waste, packaging material, stock and equipment of whatever nature which are utilised in connection with such business, unless prior written approval exempting them from this provision.

The Food Safety Authority of Ireland (2016) also states that suitable means of drainage must be provided for wastewater from basins and sinks. Where there is no access to the public drainage system, wastewater must drain into a suitable container of adequate size, and which can be carried to a discharge point without spillage. Open buckets and refuse bins are not suitable and wastewater must not be discharged onto the ground (FSAI, 2016). In a study conducted in Dhaka by Siddiqua (2016), toilets were not available nearby in several cases, thus forcing the vendors to eliminate their body wastes in nearby areas and return to their vending sites without washing their hands. Environmental

conditions and practices like this often led to the contamination of cooked food (Siddiqua, 2016). These circumstances also highlight a need for food handlers' knowledge about correct and hygienic practices when handling food.

The environment under which street foods are being prepared, vended, and consumed predisposes foods to recontamination and cross-contamination from environmental pollutants such as airborne chemicals in dust as well as exhaust discharges from moving vehicles and industrial engines and burning fumes. In addition, there may be offensive smell from accumulated waste. Moreover, the stalls that are in close proximity to the industrials waste may be affected by the effluent from industrial discharge (Alimi, 2016). The street and market vendors particularly in the African cities, identified the lack of waste removal and sanitation services as an infrastructure deficit that affects their ability to generate sales (Roever, 2014). Roever (2014) also reported that in Accra, gutters running at ground level through the markets become clogged with rubbish because of the lack of services, causing water to back up and create stagnant pools that breed mosquitoes.

Omemu and Aderoju (2008) stated that the provision of basic infrastructure and services, such as adequate supply of clean water and electricity, waste disposal services, good drainage, and toilets made available by government, is crucial in ensuring effective food safety. In developing countries, there are difficulties related to the production of food that is not only 'safe' but of good quality because many of those producing and handling food for public consumption lack the knowledge of what is required (FAO/WHO, 2005).

Vendors who are often poor and do not have a formal education, lack proper understanding of food hygiene (Mukherjee *et al.,* 2018). A study conducted in Bo City in the Southern Province of Sierra Leone by Lamin (2017) indicated that 93.1% of the street vendors did not have any knowledge or training, while only 6.9% did. More specifically, they were unaware of the importance of hygiene in the handling and storage of food, and of modern practices which ensure that the production, processing, handling and sale of food conform to health standards (Lamin, 2017). It is crucial that governments, food handlers, consumers and other stakeholders cooperate in assuring food safety from farm-to-fork (WHO, 2015). Other critical points involve hygienic practices utilised during the transport of products to the vending site and the preparation of mixed ingredients, as well as cooking, storing, serving, sanitising and waste management (Food and Drug Administration, 2014; Cortese *et al*., 2015).

The construction and layout of the food stall should ensure that any food exposed is adequately protected from the elements, from airborne dust, birds, and insects, and from members of the public. If located outdoors, this will normally mean that the food stall should be covered over and screened at the sides. The suitability of the design and construction will also depend on whether the food stall is trading indoors or outdoors as well as on the season.

1.4 Food handlers' practices

Food handlers play a significant role in making sure that food is safe throughout the chain of food production, processing, storage, and preparation. If food handlers mishandle food and disregard hygienic measures, this may introduce pathogens to food and in some cases the pathogens may survive and proliferate in adequate numbers to cause infection in the consumer (WHO, 2018). Most food vendors do not maintain the proper food-handling practices. In addition, food handlers' obliviousness about the time of spoiling, proper storage conditions and protecting foods from open-air exposure easily leads to cross-contamination of food items (Amin, 2015).

Hilario (2015) reported that most of the vendors did not use aprons, handled food with their bare hands, had long and dirty nails which were not polished and had uncovered hair. All the vendors handled money while serving food and most of them wore jewellery and other accessories. Physical hazards may be introduced by food handlers wearing jewellery, bandages or by careless food handling practices (WHO, 1989). These items can also introduce microbes in food. Personal hygiene is vital to ensure the production of safe food for consumers (Mjoka *et al.,* 2017). Food handlers should always wash their hands at every stage of food production, particularly before handling food, after eating, touching contaminated materials, using the bathroom, smoking, sneezing, coughing, handling money, and engaging in activities that may contaminate food (Mjoka *et al.,* 2017). Improper food handling can lead to the transfer of pathogens such as *Salmonella*, *E. coli* and *S. aureus* from the human body and environment into foods (Rane, 2011).

Utensils and equipment may also lead to microbial cross-contamination of food with *S. aureus, E*. *coli* and *Shigella* owing to contaminated water, dishcloths and handlers (WHO, 1996).

The hands of some of the employees in the food service can serve as a transmission route in the spread of foodborne diseases either because of poor personal hygiene or because of cross-contamination. Such a food handler may contaminate his or her hands when using the toilet and not washing hands afterwards, or bacteria might be spread from raw meat to green salads at the hands of the food handler (Kariuki, 2018). Mishandling food and disregarding hygienic measures on the part of the food vendors may enable pathogens to transfer to food and in some cases to survive and multiply in sufficient numbers to cause illness in the consumer (Akter, 2016).

Furthermore, food can also be contaminated owing to unconscious body habits of the food handler such scratching the nose or head and licking fingers, as 45-50% of the population is estimated to carry *S. aureus* and *E. coli* on the hands, nose and in the hair (Shilenge, 2014). Some food handlers may introduce biological hazards by careless food handling practices, for example, by cross-contamination after handling raw materials when they suffer from specific diseases and physical hazards. Most of the vendors pack the food in polythene bags for their customers. When packing these foods, they blow air into the polythene bags to open them. In this way several pathogens can be passed on to the consumer (Akter, 2016). In the preparation and sale of food, food handlers should

refrain from unhygienic and unsightly practices, such as touching their mouths, tongues, noses or eyes (WHO, 1996).

Some street food vendors use leftover perishable raw materials for preparation next day without a proper storage facility (Siddiqua, 2016). A study in Colombia revealed that a group of more than 30% food handlers examined, were carriers of pathogenic microorganisms, including *Salmonella typhii, S. aureus, Salmonella enteritidis,* and *Shigella* (Kariuki, 2018).

Several foodborne disease outbreaks have been determined to be a result of poor food handling practices, such as the cross between raw and cooked products, temperature abuse and the poor personal hygiene of food handlers i.e. failure to wash hands after using the toilet (Todd *et al*., 2007). In a study conducted by Bryan (1988), it has been observed that the same knife was used to cut raw meat and poultry as well as gravy and salad without being cleaned first.

Food handlers' hands and fingernails as well as aprons play a crucial role in microbial transfer amidst meat and meat products as well as equipment (Shilenge, 2014). Therefore, food handlers should wear clean and proper clothing according to prevailing local standards (Shilenge, 2014). However, the requirement for them to wear aprons of a particular colour or shade or to wear hair coverings should be tempered by the realisation

that it has more to do with food aesthetics and inspiring consumer confidence than food safety (WHO, 1996; Kariuki, 2018).

Most street food vendors undergo little or no training and lack the required knowledge on food safety and hygiene (Mafune, 2016). Sometimes food handlers may have basic knowledge regarding personal hygiene, but they do not understand the essential aspects of hygiene when it comes to food (Gadi *et al*., 2013). This makes them develop their own way of handling foods which they perceive to be hygienic and conventional (Githaiga *et al*., 2012). When food handlers mishandle food and disregard hygienic measures, pathogens may be introduced into food and in some cases the pathogens may survive and proliferate in adequate numbers to cause infection in the consumer (WHO, 2018; Kariuki 2018).

Given the challenges as highlighted, food handlers should be empowered with skills and knowledge necessary to enable them to handle food hygienically since they play a significant role in making sure that food is safe throughout the chain of food production, processing, storage and preparation (Adimasu *et al.*, 2016). The role of food handlers, especially the food vendors, in effectively reducing the risk of foodborne diseases is important as they are in direct contact with the consumers. In addition, they are the least challenging in terms of implementing food safety control measures (Iwu *et al*., 2017).

In many neighbourhoods of Johannesburg, many popular street foods are being sold by vendors who are often not regulated (Balkaran *et al.*, 2014). Nonetheless, consumers of street foods have entrusted their safety to these vendors, who cannot guarantee the implementation of food safety practices in their activities (Balkaran *et al.*, 2014). The fact that the foods are 'ready-to-eat' (RTE) means that under normal circumstances the consumer does not subject the food to further treatment that could serve to reduce levels of contamination prior to consumption (Hilario, 2015).

Furthermore, WHO (2014) has reported that not all food handlers and consumers understand their own roles in applying basic hygiene and sanitation practices when buying, selling, and preparing food (WHO, 2014). Therefore, it is important that consumers' knowledge, attitudes and behaviour towards food safety be taken into consideration by researchers to reach a full understanding of the alteration in their behaviour and beliefs necessary to help them make informed decisions about food handling and consumption (Asiegbu, 2015).

1.5 Foodborne pathogens associated with food preparation practices

Foodborne pathogens, which include bacteria, viruses, fungi and parasites, are the causative agents of foodborne diseases (Zhao *et al*., 2014). They are responsible for numerous cases of infectious gastrointestinal diseases each year, costing billions of dollars in medical care facilities (CDC, 2011). Emerging foodborne pathogens and

foodborne diseases are likely to occur driven by factors such as pathogen evolution, changes in agricultural and food manufacturing practices, and changes to the human host status (CDC, 2011). Foodborne diseases are infectious and are mostly caused by bacteria, viruses, parasites and food toxins contracted while consuming contaminated food and drink.

Pathogens present in street foods come from different sources and produced using various practices (Rane, 2011) which could compromise food safety. Improper food handling can lead to transfer of pathogens such as *Salmonella, E. coli* and *S. aureus* from human body and environment into foods (Rane, 2011). In addition, improper waste disposal has been associated with transmission of enteric pathogens such as *Salmonella, Shigella* and *E. coli*. While contaminated water has been associated with pathogens such as *E. coli,* faecal streptococci*, Salmonella* and *Vibrio cholera* while vegetables and spices are associated with introduction of spore formers such as Bacilli and *Clostridium* and pathogens such as *L. monocytogenes, Shigella* and *Salmonella* (Rane, 2011). Furthermore, improper storage temperature and reheating of food have been associated with production of heat-stable toxins produced by pathogens such as *C. perfringens* and *B. cereus* (Rane, 2011). Serving utensils used at the vending site are often contaminated with *Micrococcus* spp. and *S. aureus*. These species may have originated from the vendors' hands when they touched the food preparation areas, dishcloths, or the water during dishwashing or hand washing. This indicates cross-contamination between dishwater, food preparation surfaces, and the food itself (Rane, 2011). Barro *et al.* (2006)

reported that bacteria from dirty dish-washing water and other sources adhered to the utensil surface and constituted a risk during the food vending process. Microbiological analysis of utensils surface and knives showed the presence of *Salmonella* and *Shigella* (Barro *et al*., 2006).

Nevertheless, pathogens only represent a small portion of the food microbiota. Most foodborne microorganisms usually do not cause human diseases and are referred to as commensal bacteria (Xiaojing, 2009). While some commensal bacteria cause food spoilage, which is one of the greatest burdens to the food industry; others are of great economic value by playing an integral role in food fermentation (lactic acid bacteria), food preservation (lactococci) or functional food production (probiotics). For example, the total count of commensal bacteria could be high in fermented food. Though commensal bacteria are generally considered safe, some of them, such as *Pseudomonas, Enterococcus*, and *Staphylococcus*, are opportunistic pathogens and can cause disease to immuno-compromised individuals (Xiaojing, 2009).

Ready-to-eat meat products include high heat-processed and low heat-processed uncured and cured meat products (Oguttu, 2015). High heat-processed cured and uncured meats are given heat treatment to make them commercially sterile. Thus, they may only have some thermophilic spores surviving which will not germinate unless there is temperature abuse during product handling. Low heat-processed uncured meats, such as roasts, are given heat treatment ranging from 140 to 150°F internal temperature (60

to 65°C) for one hour or more, depending upon the size of the meat. Under this condition, only the spores of *Bacillus* and *Clostridium* spp. and some extremely thermoduric species such as *Lactobacillus viridescens* and some *Enterococcus, Micrococcus* can survive. Many other types of microorganisms can enter the products from equipment, personnel, and air as post-heat contaminants (Oguttu, 2015).

Moreover, Imathiu (2017) reported that foodborne bacterial pathogens commonly detected in street foods in developing countries include *B. cereus, C. perfringens, S. aureus* and *Salmonella* spp. These pathogens, among others, may result in foodborne infections and intoxications once contaminated food is ingested by the unsuspecting consumers. Street foods are often prepared by hand which may lead to an increased incidence of contamination with the potential foodborne pathogens (Imathiu, 2017).

Pathogenic microorganisms usually originate from an infected host (either human or other animal), or directly from the environment. Many human pathogens can only be transmitted by direct or close contact with an infected person or animal (Gerba 2015). This is because their survival time outside the host is very brief. Pathogens transmitted through the environment may survive from hours to years outside the host, depending on the organism and the environment (Gerba 2015). Pathogens may exit a host in respiratory secretions from the nose and mouth, or be shed on dead skin or in feces, urine, saliva or tears. Thus, they may contaminate the air, water, food, or inanimate objects (Gerba 2015). When contaminated air is inhaled or food consumed, the organisms are effectively

transmitted to another host, where the infection process begins again. Airborne transmission can occur via release from the host in droplets (i.e., coughing) or through natural (surf at a beach) or human activities (cooling towers, showers) Some organisms may be carried great distances, hundreds of meters or miles (e.g., Legionnaires' disease and foot-and-mouth disease) (Gerba 2015).

1.5.1 *Staphylococcus aureus*

Staphylococcus aureus is a spherical bacterium (coccus) which, on microscopic examination, appears singly, in pairs, or in bunched, grape-like clusters (Dilbaghi *et al.,* 2007). They are Gram-positive, facultative anaerobes, but grow rapidly under aerobic conditions. *Staphylococcus* is ubiquitously distributed in nature and known to be the normal flora on the skin, hair and mucous membrane of both humans and animals (Shilenge, 2014). However, *Staphylococcus* spp. can cause infections when they are introduced to sterile parts of the body. It is for this reason they are considered agents of opportunistic diseases in animals and human (Shilenge, 2014). They can be present in infections such as cuts, wounds, abscesses, and facial acne. The contamination generally occurs through these sources because of improper handling by infected persons (Dilbaghi *et al.,* 2007). In food, *Staphylococcus* has previously been isolated from a variety of foodstuffs, including meat products (Shilenge, 2014).

S. aureus remains a major cause of foodborne diseases because it can easily contaminate food products during preparation and processing (Oguttu, 2015). Furthermore, these organisms can survive on hands, knives, chopping boards and dish

cloths for hours to days following initial contamination (Oguttu, 2015). Staphylococcal food poisoning is a worldwide public health problem (Oguttu, 2015). One of the major problems associated with the control of *S. aureus* food poisoning is the high carriage rate of the organism in humans, which accounts for the high-risk contamination of food by the food handlers (Oguttu, 2015). Staphylococcal food poisoning is a gastrointestinal illness caused by eating foods contaminated with toxins produced by the bacterium *S. aureus* (Agbo *et al*., 2016). The foodborne illness develops in people who ingest food that has been improperly stored or cooked (particularly food such as salad, ice cream, ham, processed meats, chicken, pastries, and hollandaise sauce) and in which *S. aureus* has grown (Agbo *et al*., 2016). For this reason, it is advisable that the street vendors cook their food thoroughly to avoid contamination by *S. aureus* which could lead to staphylococcal food poisoning. Moreover, *S. aureus* is highly vulnerable to destruction by heat treatment and nearly all sanitising agents. Therefore, the presence of this bacterium or its enterotoxin in processed foods or on food equipment is generally an indication of poor sanitation (Barron *et al*., 2006). This indicates that interventions to resolve the challenge should focus on strengthening food-handlers' practices.

1.5.2 *Bacillus cereus*

Bacillus cereus is a Gram-positive, facultative aerobic spore former whose cells are large rods and whose spores do not swell the sporangium situated centrally or sub-terminally. It causes two kinds of foodborne disease, namely an intoxication due to a toxin preformed in the food and an infection due to the ingestion of cells which produce enterotoxins in

the small intestine (Dilbaghi *et al*., 2007). *B. cereus* is widely distributed in nature and is readily isolated from soil, dust and vegetation. Low levels of *B. cereus* cells or spores are found on virtually every raw agricultural commodity, e.g., herbs, spices, vegetables, milk and meat. These levels are generally too low to cause foodborne poisoning; however, the ability of *B. cereus* to form spores ensures its survival through all stages of food processing and subsequent time or temperature abuse enables low levels of *B. cereus* to multiply to dangerous levels (FSAI, 2016). Almost all types of foods have been associated with *B. cereus* food poisoning; however, most cases have been linked to heat-treated foods that have been subjected to temperature abuse during storage and handling, e.g. cooked rice. Temperature abuse can result in spore germination and multiplication of the vegetative cells, leading to hazardous levels of vegetative cells or toxins in the food at the time of consumption (FSAI, 2016). It is advisable that street vendors keep the food at the correct temperature to prevent the growth of contaminating bacteria. Keeping raw meats at room temperature is a dangerous practice that leaves the food open to the spread of a multitude of different bacteria (Hilario, 2015). Additionally, the ingredients used during preparation should be closed and kept away from the dust after use.

Bacillus cereus food poisoning is the general description, although two recognised types of illness are caused by two distinct metabolites (Dilbaghi, 2007). The diarrheal type of illness is caused by a large molecular weight protein, while the vomiting (emetic) type of illness is believed to be caused by a low molecular weight, heat-stable peptide. The symptoms of *B. cereus* diarrheal type food poisoning are the onset of watery diarrhoea,

abdominal cramps, and pain after six to 15 hours of consumption of contaminated food. Nausea may accompany diarrhoea but vomiting (emesis) rarely occurs. The emetic type of food poisoning is characterised by nausea and vomiting within 30 minutes to six hours after consumption of contaminated foods. Occasionally, abdominal cramps or diarrhoea, or both, may also occur (Dilbaghi *et al.*, 2007). For both types of food poisoning the food involved has usually been heat-treated, and surviving spores are the source of the food poisoning. *B. cereus* is not a competitive microorganism but grows well after cooking and cooling (<48°C). The heat treatment will cause spore germination, and in the absence of competing flora, *B. cereus* grows well. *B. cereus* is a common soil saprophyte and is easily spread to many types of foods, especially those of plant origin, but is also frequently isolated from meat, eggs and dairy products (Granum *et al*., 1997). Since *B cereus* spores can germinate when exposed to heat or improper handling, the 2013 Food Code recommends that hot foods be maintained at a temperature of 135ºF (57ºC) or above while cold foods be maintained at a temperature of 41ºF (5ºC) or below (Food and Drug Administration , 2013a). Steaming under pressure, roasting, frying, and grilling foods will destroy the vegetative cells and spores if temperatures within foods are ≥ 145ºF (63ºC) (FDA 2013a). Street vendors are advised to adhere to these temperatures recommended by the FDA to ensure that the food is not contaminated by *B. cereus* by which could lead to food poisoning.

1.5.3 *Salmonella*

Salmonella is a rod-shaped, Gram-negative, non-sporulating, facultative anaerobic motile bacterium (Dilbaghi *et al*., 2007). *Salmonella* are natural inhabitants of the gastrointestinal tracts of animals, birds, pets, frogs, turtles, and insects (Dilbaghi *et al*., 2007). They are also present in soil, water and sewage contaminated with faecal matter from where they can contaminate foods directly or indirectly and can cause Salmonellosis (Dilbaghi *et al*., 2007). Foods generally associated with outbreaks of *Salmonella* include raw meats, poultry, eggs, milk and dairy products, fish, shrimp, frog legs, coconut, sauces and salad dressing, cake mixes, cream-filled desserts, and toppings, dried gelatine, peanut butter, cocoa, and chocolate. (Dilbaghi *et al*., 2007). The important route of transmission of the *Salmonella* organism from animals to man is via food products of animal origin which may be contaminated at the source or during handling (Hassan, 2018).

According to Centres of Disease Control and Prevention (2016), *Salmonella* is a bacterium that makes people sick. Salmonellosis is a major health problem that prevails in South African regions where broiler farms and slaughterhouses have been regarded as major sources of contamination (Akhtar *et al.*, 2014). *Salmonella* is an important bacterial genus which causes one of the most common forms of food poisoning worldwide (Akhtar *et al*, 2014). It is one of the most extensively studied bacterial species in terms of its physiology, genetics, cell structure, and development. It is also one of the most extensively characterised bacterial pathogens and is a leading cause of bacterial gastroenteritis. *Salmonella* can cause a variety of disease syndromes, namely enteric

fever, bacteraemia, enterocolitis, and focal infections (Akhtar *et al*., 2014). According to Food Safety Management (Undated), *Salmonella* contamination commonly results from raw food coming into contact with ready-to eat products. Raw foods such as meats are often unavoidably contaminated with *Salmonella* which, in the normal course of events, die during the cooking process (Akhtar *et al*., 2014). For this reason, it is important that street vendors keep the raw food separate from cooked food to prevent contamination by *Salmonella*. Additionally, *Salmonella* mainly arises from faecal contamination. Therefore, street vendors are advised to wash their hands thoroughly with soap after using the toilet (Akhtar *et al*., 2014).

Salmonella is capable of infecting a large variety of both cold- and warm-blooded animals (Gerba 2015). Typhoid fever, caused by *S. typhii*, and paratyphoid fever, caused by *S. paratyphii*, are normally found only in humans, although *S. paratyphii* is found in domestic animals on rare occasions. In the United States, salmonellosis is due primarily to foodborne transmission, due to bacteria that infect beef and poultry and are subsequently capable of growing in these foods. Salmonellosis is the leading cause of foodborne illness in the United States (Gerba 2015). Since the route of transmission is fecal–oral, any food or water contaminated with feces may transmit the organism to a new host. Zishiri *et al.* (2015) reported a 51% prevalence of *Salmonella* isolated from commercial chickens and human clinical isolates. The presence of *Salmonella* species at farm level is a serious concern because it shows that the potential exists for the pathogens to disseminate from the farms to communities. This is of great concern because the majority of South Africans

depend on food sold in informal retail outlets where hygienic conditions are questionable and promote the accumulation and proliferation of pathogens, posing a danger to consumers (Zishiri *et al*., 2015). The presence of *Salmonella* in chicken meat is quite serious because South Africa imports a significant amount of poultry products from other countries such as Brazil, China and the USA. Consumers are therefore at risk of contracting salmonellosis from either local or imported poultry products (Zishiri *et al*., 2015).

1.6 Antimicrobial resistance

Studies have indicated that the prevalence of antimicrobial resistance among foodborne pathogens has increased during recent decades (Oladipo *et al.*, 2010; Eromo *et al.*, 2016). Antimicrobial resistance is the ability of microbes to grow in the presence of a chemical (drug) that would normally kill them or limit their growth. Antimicrobial resistance makes it harder to eliminate infections from the body as existing drugs become less effective. As a result, some infectious diseases are now more difficult to treat than they were just a few decades ago. As more microbes become resistant to antimicrobials, the protective value of these medicines is reduced (NIH, 2009). Overuse and misuse of antimicrobial medicines are among the factors that have contributed to the development of drug-resistant microbes (NIH, 2009).

Antimicrobial resistance emerged first in nature as organisms developed ways to survive antimicrobial production (FDA, 2013). The development of resistance by bacteria to antibiotics and similar drugs called antimicrobials is considered a major public health threat by the Food and Drug Administration (FDA) and its counterparts around the world (FDA, 2013). Resistance to a given antibiotic may be an inherent characteristic of the organism or it may be acquired (Dey *et al.,* 2018). Mutations in the bacterial genome or acquisition of additional genes coding for a resistance mechanism leads to the acquired resistance against an antimicrobial agent. The defensive mechanism of the bacteria is altered leading to the change in membrane permeability, active transport of antibiotics, enzymatic inactivation of antibiotics, target modification and routing metabolic pathways around the disrupted point owing to these genetic changes (Dey *et al*., 2018). Antibiotics play a vital role in the management of bacterial infections, reducing morbidity and preventing mortality. They are estimated to increase life expectancy by 20 years (DOH, 2014). However, the extensive use of antibiotics has resulted in drug resistance that threatens to reverse the life-saving power of these medicines. In Europe, 25 000 patients die annually from resistant bacterial infections, and in South-East Asia one child dies every five minutes from a resistant bacterial infection (DOH, 2014).

Multi-drug resistance of foodborne microorganisms aggravates the food safety situation. Selective pressure towards resistant bacteria has been created by the possibility of horizontal transfer of resistance determinants and the over-use and misuse of antibiotics. Due to these factors, the severity of the problem is significantly increased (Dey, 2018).

The intimidating probability is that most pathogenic bacteria threatening human health will soon be resistant to all known antibiotics (Dey, 2018). Once antimicrobial-resistant bacteria have become established in an animal, they may persist for long periods after the antimicrobial has become undetectable in the animal's body. Foodborne transmission of antimicrobial-resistant bacteria from food animals to people is well documented (Codex Alimentarius Commission, 2018; WHO, 2018). There are also other potential sources of antimicrobial-resistant bacteria in food such as contaminated water used to irrigate or wash food (EFSA, 2015). Food workers who carry antimicrobial-resistant bacteria may also have the potential to contaminate food with these bacteria through poor handling. Contamination of the food processing or food preparation environment with antimicrobial-resistant bacteria represents another potential source. Thorough cooking is as effective in destroying antimicrobial-resistant bacteria as it is in destroying 'wild' type bacteria. However, not all foods are cooked, and cooking is not always thorough (EFSA, 2015).

Antibiotic-resistant bacterial species observed in raw food material at retail level reportedly support the view that resistance could be transferred to humans through the food supply, although the extent to which resistant bacteria could survive under antibacterial treatment such as cooking is not fully clear (Zhao, 2009). When ingested by consumers, the antibiotic-resistant (ART) bacteria detected from ready-to-eat foods enters the intestinal tract of consumers and exchanges AR genes with the intestinal colonized microbiota of the consumer (Zhao, 2009). Therefore, the prevalence of ART bacteria in ready-to-eat food may represent more directly the real exposure of humans to

ART bacteria through daily consumption of conventional food. To date, ART bacteria have been detected in many different categories of ready-to-eat foods (Zhao, 2009).

The incidence of foodborne diseases is increasing rapidly owing to unhygienic methods of processing food (WHO, 2015). Most microbial etiological agents of foodborne diseases have developed resistance to commonly used antibiotics (CSPI, 2013). Foodborne antibiotic-resistant pathogenic bacteria such as *Campylobacter jejuni, Bacillus cereus, Clostridium perfringens, Escherichia coli, Salmonella enterica, Staphylococcus aureus, Vibrio cholerae, and Vibrio parahaemolyticus* can adversely affect both animal and human health (Friedman, 2015). In developing countries antimicrobial resistance occurs owing to an increased and indiscriminate use of antibiotics in food animals, environments and humans (Islam *et al*., 2016). In recent times, the multi-drug resistance of *Salmonella spp*. has increased alarmingly (Hassan *et al.,* 2018). It is presumed that the extensive use of antibiotics, especially in livestock production, may have resulted in the increasing incidence of antibiotic resistance in foodborne *Salmonella spp*. and other microorganisms (Hassan *et al*., 2018).

Antimicrobial resistance (AMR) has been described in many different species of bacteria (Agyare *et al*., 2018). For example, Methicillin-resistant *Staphylococcus aureus* (MRSA), now known as a superbug, is resistant to almost every available antibiotic. Beta-lactams (β-lactams) were considered the first line of drugs for the treatment of staphylococcal infections but owing to the emergence of high level of resistance to these and other drugs,

there are currently very few drugs available for the treatment of these infections (Agyare *et al*., 2018). A study in Benin revealed that 15.18% of *S. aureus* stains isolated from street foods were resistant to methicillin (Eromo, 2016).

Additionally, medically significant *Bacillus* species such as *B. anthracis* cause anthrax and *B. cereus* causes food poisoning (Agyare *et al*., 2018). *B. anthracis*, the causative agent of anthrax, is spore forming and a toxin producing rod-shaped bacterium that is classified as a category A bioterror agent (Head *et al*., 2016). This pathogenic microbe can be transmitted to both animals and humans. Clinical presentation depends on the route of entry (direct contact, ingestion, injection or aerosolization) with symptoms ranging from isolated skin infections to more severe manifestations such as cardiac or pulmonary shock, meningitis, and death. To date, anthrax is treatable if antibiotics are administered promptly and continued for 60 days. However, if treatment is delayed or administered improperly, the patient's chances of survival are decreased drastically (Head *et al*., 2016). *Bacillus cereus* is typically resistant to penicillin and other β-lactam antibiotics and can furthermore acquire resistance to commonly used antibiotics such as ciprofloxacin, cloxacillin, erythromycin, tetracycline, and streptomycin (Fielder *et al*., 2019). Foodborne illnesses associated with *B. cereus* group strains seldomly need to be treated with antibiotics. However, it has not been well investigated to which degree *B. cereus* group-strains can serve as a source of transferable antibiotic resistance genes in the food chain (Fielder *et al*., 2019).

Other infections caused by *Bacilli* spp. include pneumonia, endocarditis and, ocular and musculoskeletal infections. Antibiotics usually used for *Bacillus* infections include vancomycin, imipenem, ciprofloxacin, gentamycin, tetracycline, chloramphenicol, clindamycin and erythromycin. Most *Bacillus spp*. have been found to be resistant to broad spectrum cephalosporins and ticarcillin-clavulanate (Agyare *et al*., 2018).

According to the WHO report on global surveillance of antimicrobial resistance, the ability to treat common infections in the community and hospitals is at risk owing to the antibiotic-resistance observed globally. This is not a prediction for the future but is happening right now (WHO, 2014). Drug-resistant strains, including sulfonamide-resistant *Streptococcus pyogenes*, initially appeared in military hospitals in the 1930's and penicillin-resistant *S. aureus* began to appear in London civilian hospitals in the 1940's (Levy *et al*., 2004). Ever since then, antibiotic resistance has spread among almost all pathogens at an accelerating speed. The WHO (2014) estimated that infections accounted for 45% of deaths in Africa and South-East Asia and that these diseases were responsible for 48% of premature deaths worldwide (WHO, 2014).

In South Africa, four major outbreaks of AR have been identified at the national level. The National Institute for Communicable Disease12 reported a methicillin-resistant *Staphylococcus aureus* in 2010 (Naidoo *et al*., 2013) an extended-spectrum β-lactamase (ESBL) producing *Klebsiella pneumoniae* from 2010 to 2012 (Bamford *et al*., 2011) vancomycin-resistant Enterococci in 2012, and carbapenemase producing Entero-

bacteriaceae (Mendelson *et al*., 2015). In response to the rise in outbreaks of MDR-bacterial infections in South Africa, various organisations have been established in an attempt to curb the ABR situation. For example, the South African Antibiotic Stewardship Programme (SAASP) consists of experts in various healthcare sectors who aim to promote the prescription of appropriate antibiotics and provide education on the matter (Mendelson *et al*., 2015)

Additionally, South African government established the South African Antimicrobial Resistance Strategy framework to combat the spread of AR. Its strategies focus mainly on ARB in clinical and healthcare settings. Currently, there is no strategy in place to contain and track the movement of ARB, antimicrobial-resistant genes (ARGs) and antimicrobial-resistant gene determinants (ARGDs) of environmental isolates in an attempt to prevent them from reaching clinics and healthcare settings. Although ARGs are regarded as emerging environmental contaminants, environments have globally been portrayed as one of the main contributors to the burden of AR (Czekalski *et al*., 2014).

Issues related to AR and its magnitude in Africa are hampered by lack of a surveillance system (Ndihokubwayo *et al*., 2013). Many African reviews suggest the need for a proper continental surveillance system to gather complete and adequate data on the true extent of the AR problem. The lack of an efficient ABR surveillance system in South Africa makes it difficult to obtain data on the quantities of antimicrobials that are sold by veterinary pharmaceutical companies, indicating that a good surveillance programme needs to be

established that would aid in administering and tracking antimicrobials throughout the country. Eager *et al*. (2012) noted that resistant *S. aureus* mastitis is a current problem in the South African farming industry, followed by increased resistant *E. coli* and Enterococcus species to tetracyclines, fluoroquinolones, sulphonamides, amoxicillin, and trimethoprim-sulpha combinations. In addition, high resistance in *S. aureus, Campylobacter jejuni,* and some *Listeria* and *Salmonella* species has been reported for tetracycline from a poultry abattoir. High resistance was also reported in *S. aureus* isolates for penicillin and amoxicillin in cattle that had mastitis (Henton *et al*., 2011).

Additionally, it was found that antibiotic usage in the agriculture industry has been increasing. Growth promoters that are currently being used in South Africa are banned in other countries (Moyane *et al*., 2013). Consequently, South Africa has recently been considered a major contributor to the increase in antibiotic use worldwide (Mendelson *et al*., 2015)

The main threat of ABR to human health is the transfer of resistant bacteria between animals and humans. Case studies have reported evidence of resistant *Campylobacter, Salmonella, MRSA* and *E. coli* causing diseases in humans that originated from animals (Phillips *et al*., 2004; Bengtsson *et al.,* 2014). It is alarming that the same classes of antibiotics are used for veterinary and human medicine, namely β-lactams, sulphonamides, tetracyclines, macrolides, lincosamides, streptogramins, and quinolones (Phillips *et al*., 2004). Efforts should focus on minimizing the transmission of food-borne

pathogens via the food chain, despite their antibiotic susceptibility profile by adopting good hygiene practices at all stages of food production, including food marketing and food preparation by the consumer (van den Honert, 2019).

1.7 Rationale

Street foods are perceived to be a major public health risk owing to a lack of basic infrastructure and services to support the trade. In addition there is difficulty in controlling the large numbers of street-food vending operations because of their diversity, mobility, and temporary nature (Rane, 2011). Street foods are also a potential source of foodborne illness, thus posing a major health problem that must not be overlooked (Mukherjee, 2018). Although governments throughout the world attempt to improve the safety of the food supply, the occurrence of foodborne disease remains a significant health issue in both developed and developing countries (WHO, 2010). The WHO (2015) has highlighted the challenges associated with food safety under the slogan “From farm to plate, make food safe” and emphasised various ways that can be implemented to make food safe in different environments.

Consuming unsafe foods poses a significant public health threat in the African region. For example, infants, young children, pregnant women, the elderly and those with underlying illnesses are particularly vulnerable (WHO, 2015). These unsafe foods have placed a major strain on health systems and have hurt national economies, development and

international trade in the African region (WHO, 2015). Although antimicrobials, such as antibiotics, are essential to treat infections caused by bacteria, their overuse and misuse has been linked to the emergence and spread of resistant bacteria, rendering the treatment of infectious diseases ineffective. As a result, antimicrobial resistance is increasingly becoming a bigger threat to the effectiveness of modern medicine (WHO, 2015).

In countries where street-food vending is prevalent, there is commonly a lack of information on the incidence of foodborne diseases related to street foods (WHO, 2018). Foodborne infections may lead to hospitalisations and death (Dey *et al*., 2018). Unfortunately, outbreaks of foodborne diseases are not always recorded on medical databases since their symptoms are often moderate and self-timing. Affected consumers may not even visit doctors (Oguttu, 2015). Consumers may not recognize that symptoms are due to foodborne disease and as a result, unhygienic or risky practices during the preparation and preservation of street food are ignored (Oguttu, 2015).

Additionally, lack of infrastructure plays a major role in the contamination of street foods. The provision of infrastructure and services is usually very expensive, and it is beyond the resources of most authorities to apply this strategy on a broad scale (WHO, 1996) in this challenge continues to exist (Rane, 2011). However, by identifying the critical infrastructure and service requirements of specific street-food vending operations and by helping to rank operations according to risk, hazard analysis and critical control points

(HACCP) can be used to target the provision of these facilities where they will be of the greatest benefit to protecting public health (WHO, 1996). It should be noted that in many countries, street food vendors are part of the social and cultural fabric of their communities and therefore an effort should be made to keep them as close to their current business sites as possible, even though some facilities may not be available. In this context it should be recognised that the relocation of vendors can reduce the number of factors contributing to foodborne disease (e.g., potable water and toilets - provided local authorities can supply potable water) but may not automatically impact on raw material contamination, cross-contamination, personnel hygiene, poor food preparation practices or hot and cold holding capacity. Consequently, relocation should not be a panacea for resolving the problems of street foods (WHO, 1996).

Sanitary facilities are rarely available for workers. Wastes are disposed of nearby, providing nutrients for flies and rodents and this practice may harbour foodborne pathogens. As a result, ready-to-eat (RTE) prepared street foods are commonly exposed to a variety of potential public health risks (Amare *et al.,* 2019). Additionally, unhygienic practices by street vendors lead to the contamination of food by various bacteria. Most of the bacteria present in foods are resistant to antibiotics and it is very difficult to control them (Sabuj *et al*., 2018).

This study therefore seeks to investigate the prevalence of antibiotic-resistant bacteria in the street food of Bloemfontein city. Interviews were carried out with the street vendors to

assess their knowledge of food safety. Additionally, the study investigated the role that factors such as inadequate waste management and water play on the safety of street foods. Lastly, an assessment of the microbiological quality of the foods sold in the streets was carried out.

1.8 Aim

The aim of this study was to investigate the presence of antibiotic-resistant bacteria in street-vended foods in the Mangaung Metro.

The overall objectives of the study were the following:

•To assess the knowledge, attitudes, and practices among street vendors in Mangaung Metro by means of questionnaires and a checklist.

•To isolate and identify bacterial samples from surfaces and foods sold by street vendors in Mangaung Metro.

•To investigate the prevalence of antibiotic resistance in bacteria isolated from surfaces and food samples collected.

In order to understand the overall aim of this study, the following aspects were considered: food contamination, environmental hygiene, food safety and food hygiene, food handlers, foodborne pathogens, and antimicrobial resistance.

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CHAPTER 2

ASSESSMENT OF KNOWLEDGE, ATTITUDES AND PRACTICES OF STREET VENDORS IN MANGAUNG METRO

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2.1 Abstract

Background: Microbial contamination of ready-to-eat foods and beverages sold by street vendors is an important public health concern. Despite interventions within the food safety field, evidence still shows that foodborne disease outbreaks are a result of incorrect food handling practices, such as the cross-contamination between raw and cooked products, temperature abuse, and the poor personal hygiene of food handlers i.e., failure to wash hands after using the toilet. Thus, this study assessed the knowledge, attitude, and practices of street food vendors.

Materials and methods: Street vendors in the city of Mangaung Metro were interviewed in order to assess their knowledge, attitudes and handling practices. A semi-structured questionnaire and checklist were used in interviews to determine the status of the vending sites and associated food-handling practices. The questionnaire was compiled in Sesotho language to ensure that the participants understood the questions clearly.

Results: The majority of respondent were black females. All (100%) participants' responses indicated that they had not undergone food safety training. However, participants showed a positive attitude towards food safety. Despite the positive attitude, vendors showed non-compliance when it comes to the handling of food. The inspection checklist revealed that vending stalls lack basic infrastructure such as toilets and potable water.

Conclusion: This study indicates a need for improvement of the environmental conditions at these sites to prevent foodborne diseases, this will require government intervention. Moreover, based on the observed practices, food safety and food hygiene training or workshops for street vendors are needed on a continuous basis.

Keywords: Street foods; food safety; food hygiene; environmental conditions; foodborne illnesses

2.2 Introduction

The term ‘street food’ refers to a wide variety of ready-to-eat (RTE) foods and beverages sold and sometimes prepared in public places. Street food may be enjoyed/consumed in the place where it was purchased or can be taken away and eaten elsewhere (WHO, 1996). A hawker or vendor sells it from a portable stall, cart, or food truck (Islam, 2016). This industry plays an important role in meeting the food requirements and needs of urban dwellers in many cities and towns in developing countries. The sector feeds millions of people daily with a wide variety of foods that are relatively cheap and easily accessible (Eromo *et al*., 2016). In many streets of developing countries, like South Africa, the sale and consumption of ready-to-eat foods and beverages is a common and normal phenomenon of everyday life (Kok *et al*., 2014).

The main health hazard associated with street foods is microbial contamination (Bereda *et al*., 2016). There is statistical evidence that shows that 70% of bacterial food poisoning is caused by caterers (Annor *et al*., 2011). This is greater than occurrences reported from any other food sector. Most of these food poisoning outbreaks are due to the inadequate time and temperature control of food, whereas the remaining 30% are because of cross-contamination (Annor *et al*., 2011). The food is sometimes prepared long before its consumption, stored at ambient temperature and undergoes inadequate cooling and reheating processes, leading to the contamination of the food (WHO, 1989).

Street vendors generally use carts and stands and they do not have easy access to running water. Furthermore dish and handwashing is done using the same bucket, sometimes without soap (Akter, 2016). Toilets are usually not available nearby. In some cases this forces the vendors to eliminate their body wastes in nearby areas and come back to the vending sites without washing their hands. Environmental conditions and practices such as these often result in the contamination of cooked food (Akter, 2016). Additionally, garbage and wastewater are usually discarded in the streets nearby, thereby attracting and providing food for rodents and insects (Akter, 2016).

In some cases, street vendors may completely ignore basic food hygiene practices, but consumers are probably not aware of how necessary it is to insist on safe food (Thanh, 2015). Chukuezi (2010) showed that majority (90%) of food vendors handle money while serving, only 24% prepare food in a hygienic and safe environment, while 42.7% were not wearing an apron, 48% handled food with unprotected hands (without gloves) and 52.4% did not cover their hair. Generally, 28.6% puffed air into plastic bags before use and 19.1% wore jewellery while serving foods. These are a cause for concern. The Safety Food Authority of Ireland (2016) states that jewellery must not be worn except for a wedding band, sleeper earrings or studs. Other reasons for the contamination of street fast foods include utensils and equipment that enhance cross-contamination, vending sites that are polluted, and lack of tap water used in the preparation of food (SFAI, 2016). The study therefore seeks to asses knowledge, attitudes and practices of street vendors in Mangaung Metro by using a semi-structured questionnaire and checklist.

2.3 Materials and methods

The survey was conducted in the Mangaung Metropolitan Municipality. This metropolitan municipality governs Bloemfontein and seven other surrounding towns in the Free State province of South Africa, which include Botshabelo, Dewetsdorp, Mangaung, Soutpan, Thaba Nchu, Van Stadensrus and Wepener (Figure 2.1). The Metro central office is located in Bloemfontein in the Free State province of South Africa. Bloemfontein is the capital city of the Free State with approximately 520,000 residents and forms part of the Mangaung Metropolitan Municipality that has a population of 747,431. It is one of South Africa's three national capitals and the seventh largest city in South Africa. It is situated at an altitude of 1,395 m (4,577 ft) above sea level.

This study was limited to Thaba Nchu, Botshabelo and Bloemfontein due to the high observed activity of vendors in these areas. During the survey, a total of fifteen (15) street vendors were interviewed in Bloemfontein, Botshabelo and Thaba Nchu. Five street vendors were interviewed from each town. Inclusion and exclusion criteria were used to select the participants. Street vendors that were situated far from the taxi ranks and who did not sell meat were excluded from the study. The inclusion criteria consisted of street vendors that sold meat and were located close to the taxi ranks where they are close to people and where the contamination risk was high since the majority of these vending sites lack basic infrastructure and services such as potable running water. Permission was requested from the municipality before the study was carried out. Moreover, street vendors were provided with consent forms to sign before the interview. These consent

forms were translated into the preferred language of the participants. Thereafter, it was explained to them that participation in the study was voluntary, that the names and personal details of the participants would not be revealed, and that all the documentation and information obtained during the study would be kept confidential. It was further explained to them that they had the right to withdraw at any time without providing a reason. The data was collected by means of a face-to-face interview. All questions were designed in English but translated into the language with which the respondents were comfortable (Sesotho).

A structured questionnaire survey for food handlers was used in the study. The questions that were addressed in the questionnaire survey included the demographic characteristics of food vendors, the food safety and hygiene knowledge of street vendors, waste management around the stalls by the vendors and the availability of washing facilities and potable water around the vending stalls. The questionnaire comprised both closed ended and open-ended questions to obtain more information from the vendors. A checklist was used to assess food safety practices of street foods vendors such as having long nails, wearing jewellery, washing hands, covering hair and wearing protective clothes. The checklist was also used to observe the environment around the vending stalls as well as to obtain information about facilities, namely the availability of potable water and toilets as well as adequate washing facilities. Environmental health practitioners (EHPs) at the Mangaung Metro Municipality were contacted and made aware of the research.

Figure 2.1: Mangaung Metro Municipality map (Municipalities of South Africa ,2020).

2.4 Results and discussion

Based on the results, Table 2.1 indicates that of all 15 participants interviewed in Mangaung Metro, the majority of the respondents were females. Seventy-five (75%) of the surveyed vendors were women and twenty-five (25%) men. The finding is in line with a study conducted in Ethiopia, which indicated that the majority (75%) of the vendors were young females (Amare, 2019). It was also observed at most of the stalls that men were the owners of the stalls and the females were only assisting with the preparation and serving of foods.

Table 2.1: Demographic characteristics of vendors

Characteristics	Demographic characteristics	Frequency (percentages)
Gender	Female	75%
	Male	25%
Preferred language	English	0%
	Sesotho	100%
Food safety courses	Yes	0%
	No	100%

Generally, reports indicate that men do not like to do the cooking, so women are left to take care of this aspect of the business (FAO, 2012). In South Africa and West Africa, reports indicate that women made up to 90.5 – 98.5% and 89-98% respectively of the people involved in the street-food vending business (FAO, 2012).

This number could be attributed to the fact that traditionally among African cultures, cooking and serving food is reserved for women, while men (FAO 2012) do manual labour. The results also indicate that the vendors (100%) did not receive food safety training which is disquieting. Iwu *et al.* (2017) also reported in a study they conducted that the majority of the respondents (68%) did not undergo any training on food hygiene. Similar findings by Lamin (2017) showed that 93.1% of the vendors did not have any knowledge and training, while only 6.9% did. To prevent street food contamination in South Africa, the Regulations Governing the General Hygiene Requirements for Food Premises and the Transport of Food (1999) published by the Department of Health state that any person working on the food premises must be adequately trained in food hygiene by an inspector or any suitable person. However, this was not observed in the current study and this might be due to the informal nature of the business.

EHPs are mandated by law to carry out an inspection at all food vending stalls in South Africa (Department of Helath, 2004). If the EHP, after having carried out an inspection, having due regard to existing conditions of the adjacent land and facilities, is satisfied that the food premises concerned, are favourable, the local authority will issue a certificate of

acceptability. From the results obtained in the study, the vendors mentioned that the EHPs last inspected their stalls few years ago. However, Iwu (2017) in a study conducted in Nigeria, reported that 46% of the respondents were visited at one time or the other by an environmental health worker. This is of concern because in the Manguang Metro, the stalls at which the street vendors are selling food are not conducive to food preparation. The stalls are not properly covered and are exposed to dust, motor vehicle fumes and insects. Dust has the potential to carry many microbes that may be pathogenic if left to settle on prepared foods.

Hence it is important that food prepared by vendors is covered to protect it from such exposure (Muinde *et al*., 2005). Additionally, this highlights the importance of monitoring the stalls as regularly as possible by EHPs to ensure that they are in a condition conducive for both vendors and consumers. In developed countries regulations such as Regulation (EC) No 852/2004 of the European Parliament and of the Council of 29 April 2004 on the hygiene of foodstuffs assert that the layout, design, construction, siting and size of food premises are to be carried out in a manner that protects against the accumulation of dirt, contact with toxic materials, the shedding of particles into food and the formation of condensation or undesirable mould on surfaces.

Responses regarding the knowledge of food safety are shown on Table 2.2. All vendors agreed that it is important to wash hands before handling food. During the observations shown on Table 2.3, the vendors did indeed wash their hands with soap before handling

food. However, all vendors surveyed handled food with bare hands. This practice might cause cross-contamination between raw and cooked products. Handling money while serving food was also observed. Hands are the most significant source of transfer for microorganisms from faeces, face, skin or other sites to vendors (Balkaran *et al*., 2014). The vendors can be carriers of pathogens such as *E. coli, Salmonella, Shigella, Campylobacter* and *S. aureus* and eventually transfer these foodborne hazards to the consumers (Rane, 2011; Balkaran *et al*., 2014). Therefore, it is prudent that cooked food should not be handled with bare hands. According to the Guidelines for the Design of Control Measures for Street-vended Foods in the Near East (2010), food should preferably be handled with clean tongs, forks, spoons or disposable gloves (FAO 2010; Samapundo, 2015).

Moreover, 99% of the vendors (Table 2.2) stated that they kept their raw meat in cooler boxes (Figure 2.2) which are separate from the cooked food. This observation is compliant with essential safety requirements for street foods (WHO, 1996) which states that an important principle in preparing and processing food is to avoid direct and indirect contact between raw and cooked or prepared foods that will be consumed without further heating. This practice by the street vendors reduces cross-contamination. During the survey, it was noted that only one vendor had a refrigerator for food storage. However, she did not know the correct temperature at which to store the food.

Table 2.2: Assessment of vendors' food safety knowledge responses

Statement No	Yes	No
1. It is important to wash your hands before handling food	**100%**	**0%**
2. It is important to wear protective clothes while cooking	100%	0%
3. Knives and cutting boards should be properly sanitized to prevent cross contamination	100%	0%
4. Raw and cooked foods should be stored separately to reduce the risk of food contamination	100%	0%
5. Do you have storage facilities (refrigerator) for both raw and cooked meat?	100% (cooler boxes)	0%
6. Do you know the correct temperature for the refrigerator?	99% (cooler boxes)	1%
7. Do you reheat your food before serving?	0%	100%
8. Food prepared in advance reduces the risk of food contamination	0%	100%
9. Dish towels can be a source of food contamination	100%	0%

Table 2.3: Food-handling practices observations

Observation	Yes	No
1. Food prepared at the stall	100%	0%
2. Wash hands with soap before cooking	100%	0%
3. Wash food before cooking	100%	0%
4. Preparation surface clean	2%	98%
5. Prepared on same surfaces more than twice	98%	2%
6. Use of apron	60%	40%
7. Long nails	1%	99%
8. Wears jewellery	2%	98%
9. Handles food with bare hands	100%	0%
10. The vendor's clothes are clean	99%	1%
11. Hair covering	99%	1%

Figure 2.2: Cooler box used to store raw food prior food preparation by street vendors.

According to the guidelines for safe street foods, a clean environment, separation of raw and cooked foods, thorough cooking and refrigeration, the use of safe water and safe raw materials and raised awareness are recommended to assure the safety of RTE foods (INFOSAN, 2010). On the other hand, vendors (100%) as indicated in table 2.2 seemed to be unaware of the importance of reheating food to prevent foodborne diseases.

If reheating is inadequate, bacterial contaminants can multiply. It was also observed that 99% of the street vendors know the correct temperature to store their raw materials; they kept them in cooler boxes with ice inside (Table 2.2). Only one vendor indicated that she did not know the correct temperature to keep meat. According to the WHO (2007), microorganisms grow and multiply rapidly at the temperature range of 5°C to 60°C. Foods thoroughly fried or cooked and consumed while hot are inherently safer than pre-cooked foods, especially when the latter are held at ambient temperatures (15-40°C) for more than four to five hours (WHO, 2007). Prepared foods served hot should be kept at a temperature of at least 60°C to prevent microbial growth, particularly if the sales period extends over four or five hours (WHO, 1996).

In terms of the legislation regarding general hygiene requirements for food premises and the transport of food, anyone preparing food should wear protective clothing such as aprons and head coverings such as nets or scarves (Campbell, 2011). Based on observations reflected in Table 2.3, responses from food handlers indicate that 99% of street vendors showed compliance with legislation by not wearing any jewellery and

keeping their nails short and clean when preparing food. However, 40% of the vendors showed non-compliance with regard to wearing aprons during food preparation and serving. Other studies have also observed that a low percentage of vendors wear aprons and gloves while handling, preparing and serving food (Chukuezi, 2010; Lues *et al*., 2006; Samapundo, 2015). Only 60% of the vendors in this study wore aprons while handling, preparing and serving food (Table 2.3). The 60% of vendors wearing aprons and head coverings were mostly women; this may be due to a cultural norm or value that requires food to be handled while wearing an apron and head covering (Campbell, 2011). Additionally, head coverings may be because of certain cultural requirements requiring the heads of married women to be covered.

Owing to a lack of space, 98% of the vendors showed non-compliance with regard to food handling. They prepared foods several times on the same surface (Table 2.3). In Figure 2.2, it can be seen that surfaces were visibly not clean. It was also observed that these surfaces had remains of food prepared earlier, which can become sources of cross-contamination. Again, an observation showed that food handlers did not sanitise working surfaces before and after food preparation (Figure 2.3). On this aspect, educational training for food handlers is needed to ensure proper cleaning and sanitising of working surfaces to ensure a minimal likelihood of contamination of food products prepared for consumers. It has been established that *Salmonella* non-typhi, *Salmonella*, *Campylobacter* and *E. coli* can survive for days on fingertips and other surfaces (Mlay, 2018).

Figure 2.3: Preparation surface of street vendors during food preparation

According to the results depicted in Table 2.4, the disposal of garbage was considered to be one of the biggest problems at the selling points in Bloemfontein. None of the vendors (100%) as indicated in Table 2.4 had trash bins; instead, they used plastic bags and boxes as trash bins. This demonstrates that even though the vendors did not have waste bins, they were aware of the importance of cleaning the environment around them.

These findings were similar to those observed by Muyanja *et al*. (2011), where the vendors were using gunny bags for the collection of waste. Street Vendors' Laws and Legal issues in South Africa (2014) indicated that one of the challenges identified are insufficient services such as waste bins. Mangaung Local Municipality By-Laws Relating to Street Trading (2006) stipulates that everyday street vendors should remove from any public road or public place at the conclusion of selling, all waste, packaging material, stock and equipment of whatever nature which are utilised in connection with such business, unless prior written approval exempting him from this provision, has been given by the "Council".

Table 2.4: Environment around the stalls

Observation	Yes	No
1. Trash bins available for customer use —they use plastic	100%	0%
2. Enough trash bins available for disposal of solid wastes	0%	100%
3. The environment around the stall is clean: far from rubbish	99%	1%
4. Presence of vectors e.g. cockroaches, rodents, around the stall	0%	100%
5. Stalls are located far from toilet facilities	100%	0%
6. Vending stall protected from sun, wind and dust	0%	100%
7. Location of the business interfere with human or traffic flow	0%	100%
8. Environmental health practitioners visitation to street vendors	0%	100%

The Regional code of hygiene practice for street foods in Asia adopted in 2017 (Bisogno, 2019) states that all solid waste should be properly disposed of in suitable containers that are secured with tight-fitting lids or placed in waste bins. Collection of garbage should be done by municipalities on a regular basis. The absence of waste bins causes concern as this might attract flies and insects that are potential vectors of pathogens. During the observations at the time of sample collection (Table 2.4) no flies, cockroaches or other vectors were visible around the stalls. It must be noted that the study was conducted during the winter season and this might be the reason for the absence of flies around stalls.

Moreover, during the study it was observed as indicated at Table 2.4 that the stalls were made of steel and covered with sailcloth sheets. The structures of these vending stalls do not provide protection from the sun and dust and further investigation must be conducted regarding these concerns. Furthermore, shelter has been particularly observed as an issue in other African cities, where the hot sun and heavy rains affected their work: vendors of perishable products had their goods spoilt, while other goods were damaged by the sun and rain (Roever, 2014). This was also observed in the current study, in which the vendors stated that they are affected by rain during the rainy season as their stalls are open and cannot protect them from the rain.

In this study, 70% of the food vendors indicated that they had water drainage systems near their vending stalls, however, some of these drains were blocked and smelly (Table

2.5). Refuse disposed of in stormwater drains may cause blockages and encourage fly and mosquito breeding. It is therefore important that waste is disposed of properly. The Mangaung Metro should also ensure that their waste and wastewater drainage systems are managed properly and checked regularly.

Table 2.5: Assessment of environment around the stalls responses

Statement	Yes	No
1. **Are dustbins available for customers use?**	0%	100%
2. **Do you have drainage systems available for disposal of liquid wastes?**	70%	30%
3. **Do you have your own waste bin, or do you share it with other vendors?**	100% (plastics)	0%
4. **Do you experience a problem of waste disposal during strikes by waste collectors?**	100%	0%

Consistent with this are the findings of Githaiga *et al*. (2012) who found that a high frequency of stalls (85%) in enterprise roads had food preparation and serving areas with drainage tunnels nearby. During the current study 30% of surveyed vendors stated that they must walk a distance to reach the drainage systems. Therefore, they keep the dirty water in the extra containers all day until the close of business when they have time to discard the water in the drains. This is a cause for concern because keeping dirty water around the stall is unhygienic and may harbour pathogens. Some street vendors expressed concern about the non-functionality of drains, causing some customers to complain about the safety of the food they buy daily.

Furthermore, the results of the survey in Table 2.6 indicate that there is no potable water supply near the vending stalls in Mangaung. It was found that 99% of the food vendors surveyed obtain water from the nearby stores. Vendors buy water every day for R3.00 per 20 litres.

Table 2.6: Water availability around the stalls

Observation	Yes	No
1. Source of water at the stall/nearby	1%	99%
2. Drainage for disposal of wastewater at the stall/nearby	70%	30%
3. Hand washing facilities available nearby	1%	99%
4. Water stored in clean containers	100%	0%
5. Water reused more than twice	100%	0%

Lues *et al*. (2006), who conducted a study in Bloemfontein, also reported that street vendors had limited access to potable water and had various means of obtaining water such as nearby shops or bringing the water with them from home. Based on these results, this suggest that no improvements have been made regarding the availability of potable water since the study conducted in 2006. According to requirements of street foods (WHO, 1996), in some countries it has been possible to locate street food vendors in specially designed centres. This grouping makes it possible to provide common facilities (potable water and electricity supplies, waste disposal services, drainage, toilets, and vehicle parking). In addition, common utensils can be centrally supplied and cleaned. The provision of such infrastructure and services is usually very expensive and it is beyond the resources of most authorities to apply this strategy on a broad scale (WHO, 1996; Al Nuaimi *et al*., 2015).

The absence of water points near their workplaces and poor drainage facilities make it difficult for vendors to practise good hygiene. In this study the vendors also revealed that in their attempts to cut costs, they buy limited quantities of water and therefore have insufficient water for washing their crockery and food stuff. As a result, food vendors reuse their water for washing utensils more than twice. That is unacceptable according to Regulations Governing General Hygiene Requirements for Food Premises and the Transport of Food, GNR. 918 of (1999). Additionally, all the vendors stated that they only change the dishwashing water when the water changes colour. This practice compromises hygiene because they believe that clean water can be determined through

observation. However, that is not true (Khairuzzaman, 2014). This perception by food handlers that clean water can be easily identified by the naked eye is a concern and may need to be further explored to determine whether this is due to the proximity of tapped drinkable (potable) water in this study area or to misconceptions about microbes. The perception that uncontaminated water may be detected with the naked eye is concerning, as with a perception like this, people may drink contaminated water based on sight, whilst exposing themselves to deadly infections such as cholera. Literature (Khairuzzaman, 2014) indicates that without enough water, hygiene and sanitary standards cannot be met. This is a major concern that must be addressed.

With regard to storage, the vendors mentioned that they do not have enough storage for their utensils and equipment. During operation hours they store their utensils (pots, cutlery, dishes) on and under a small table (Figure 2.4). Owing to a lack of storage space, they keep their equipment (chairs, tables, tents) in the storage rooms of the nearby stores. The owners of the store charge them an amount of R20 per day. Depending on how much equipment (chairs, tables, tents) they have, they might even pay more than that anticipated amount. They travel with their utensils every morning when they come to work and carry them home at the end of the day. Due to lack of storage, they end up buying less stock so that they can take leftovers home and bring them back the next day.

Figure 2.4: Vendors keep their utensils on a small table during business hours.

This is a cause for concern. According to Essential Safety Requirements for Street Food (WHO,1996), most street vendors plan food preparation in such a way that prepared, or semi-prepared items are consumed by the end of the daily business period. However, sometimes they do have leftovers. The latter is potentially hazardous food and if no cold storage (<10°C) is possible, the vendors should be encouraged to discard it. If cold storage facilities are available, the food should be kept for sale after re-heating (>70°C) on the following day. However, it is preferable to discard leftovers, especially foods liable to support microbial growth.

Moreover, only 5% of the interviewed street vendors reported that they pay monthly rent to the local municipality. However, despite their contribution towards stall rental fees, the current results have indicated that their stalls lack shelter, running water, toilets and infrastructure for waste management. These deficits make it difficult for their businesses to compete with the nearby stores. Furthermore, the respondents indicated that they believe their rent payments were not used to provide any services or infrastructure towards improving their circumstances. Mangaung Local Municipality by-laws relating to street trading (2006) stipulate that all street trading sites are leased to the street traders at a fee as determined by the Council from time to time. Street Vendors' Laws and Legal Issues in South Africa (2014) also state that a municipality may charge an annual rental fee. However, it is not clear in the legislation what this fee should be used for. Moreover, street vendors stated that they would also like to have access to electricity since this would be considerably cheaper than buying gas every week or charcoal daily.

The attempt to obtain the total number of street vendors in Mangaung Metro was in vain. The EHPs at the Mangaung Metro stated that they do not have a database where the records of street vendors are kept. This finding indicates that not all street vendors register their business with the local municipality. Mangaung Local Municipality by laws relating to street trading (2006) stipulate that no person may conduct the business of a street trader in the business area without being in possession of a valid street trading identity card issued by the “Council”. The Reginal code of hygiene practice for street foods in Asia adopted in 2017 (Bisogno, 2019), also states that street-food vendors should have registration or licensing from a relevant authority before starting their business. The registration or identification proof issued by the relevant authority should be displayed on the cart or kiosk.

EHPs also pointed out that they do not get to visit the street vendors as frequently as they should because of their tight schedules and sometimes because of the lack of resources. However, according to the guidelines of EHPs on food safety control at special event by the Department of Health South Africa (2004), activities of local authorities related to food safety control include routine monitoring of foodstuffs and food premises. This includes inspections and sampling activities as well as health education of food processors, handlers and consumers, especially within the informal sector. Lastly, their activities include advising existing and prospective entrepreneurs of requirements related to food premises and the safe handling of food this aspect is not implemented often enough as most vendors indicated lack of food safety training.

2.5 Conclusion

Despite the challenges faced by vendors, their attitude towards food safety was generally positive, although some the actual food handling practices by street vendors raised some serious concerns. Even though the vendors had a positive attitude towards food safety, non-compliance with food safety regulations was observed. Some of the vendors did not wash their hands and did not wear aprons during the processing and serving of food. The vendors mentioned that they have not received any training in food safety and hygiene. It is therefore recommended that food handlers undergo basic training in food hygiene. This is to ensure that they follow the required rules for proper hygiene and sanitation. In addition, authorities must implement policies aimed at assisting, controlling and maintaining the street food sector to ensure the safety of street food. It was observed that the material used for the construction of the stalls was not able to protect the food from dust, given that all the stalls were built on the roadsides, which were dusty and prone to fumes from cars. As stipulated by Regulations Governing General Hygiene Requirements for Food Premises and the Transport of Food, GNR. 918 of (1999), food premises should be effectively protected by the best available method against contamination or spoilage by offensive gases, smoke, dust, insects and other vectors or biological contamination. In addition, lack of storage space seemed to be an issue with all the vendors at the Mangaung Metro. Essential Safety Requirements for Street Food (WHO, 1996) also states that sales points, stationary or ambulant, should be located in a place where the risk of contamination from rubbish, sewerage and other noxious or toxic substances is absent or minimal. If such risks cannot be eliminated, food offered for sale should be suitably covered and protected from contamination. The local government should

consider the establishment of street food centres with adequate facilities and utility services at a location that will not compromise the sustainability of the vendor's businesses. Such centres will provide the necessary utilities such as potable water, adequate light, drainage, and solid and water disposal, creating a conducive environment for consumers to be served with safe food and providing a suitable setting for the relevant authorities to conduct information, education and training programmes for vendors.

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CHAPTER 3

MICROBIAL LEVELS ON STREET VENDED FOODS AND PREPARATION SURFACES AT MANGAUNG METROPOLITAN MUNICIPALITY IN SOUTH AFRICA

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3.1 Abstract

Background: The street food sector has become an important component of the food distribution system in many cities in both developing and industrialised countries, particularly for midday meals. However, certain street food can pose a significant risk to consumers because of microbiological contamination.

Materials and methods: The study was conducted in Mangaung Metropolitan Municipality during the winter season. Samples were collected through convenience sampling from the representative towns Thaba Nchu, Botshabelo and Bloemfontein. Using swabs, surface samples were collected and quantified from selective media. Eight beef samples were also collected; the microbial load on each sample was quantified and identified using a RapID kit .

Results: The surface swabs obtained in Botshabelo (1.1×10^4 CFU/m^2– 1.1×10^6 CFU/m^2) showed higher microbial counts as compared to those obtained in Bloemfontein (1.1×10^4 CFU/m^2– 1.1×10^5 CFU/m^2) and Thaba Nchu (1.1×10^4 CFU/m^2 – 1.1×10^5 CFU/m^2). Higher microbial counts were observed on meat samples sampled in Thaba Nchu (50 CFU/g x 10^5 CFU/g), Bloemfontein (48 CFU/g x 10^4 cfu/g) and Botshabelo (33 CFU/g x 10^5 CFU/g) when compared to international microbiological standards. After assessing the microbial levels, *Staphylococcus aureus, Escherichia coli, Candida guilliermondii, Corynebacterium jeikeium, Psychrobacter phenylpyruvicus* and *Peptostreptococcus tetradius* were identified.

Conclusion: This study confirmed contamination of surfaces and food served by vendors in Mangaung Metropolitan Municipality. The identified foodborne bacteria could pose a public health problem in each specific locality.

Keywords: Microbial contamination; street foods; surface swabs; food safety.

3.2 Introduction

According to the United Nations Food and Agricultural Organisation (FAO), about 2.5 billion people consume street food every day (Da Silva *et al*., 2014). Therefore, the demand to ensure the supply of safe food has been one of the major challenges and concerns for producers, consumers and public health officials globally (Da Silva *et al*., 2014). This is because foods excessively contaminated with pathogenic and spoilage microorganisms are undesirable and can cause foodborne illnesses (Hertanto et al. 2018). Transmission of pathogenic microorganisms to food takes place through inadequately washed hands and dirty clothing, such as aprons that are worn during the preparation and serving of food (Hertanto *et al*., 2018). In addition, contaminated foods can endanger public health by causing various acute and chronic foodborne illness through pathogenic microbes or toxic substances present in them (Nazni & Jaganathan 2014).

Foodborne illness, also called 'foodborne disease', 'foodborne infection' or 'food poisoning', is a common, costly but preventable public health problem (Akter, 2016). Foodborne illness can be caused by bacteria, parasites, toxins, and viruses. Amongst the common pathogens are *Salmonella* and *Escherichia coli*, which account for 52 000 and 37 000 deaths, respectively, (WHO, 2015). Each year, reports indicate that one in six Americans gets sick by consuming contaminated foods or beverages (Akter, 2016). In South Africa, a progressive increase in listeriosis cases was noted starting in mid-June 2017, heralding what was to become the world's largest listeriosis outbreak. A total of 1060 cases were reported for the period of 01 January 2017 – 17 July 2018 (Smith *et al*.,

2019). Different disease-causing microbes, or pathogens, can contaminate foods. In addition, poisonous chemicals, or other harmful substances, can cause foodborne illness if they are present in food (Akter, 2016).

The global burden of foodborne illness states that each year as many as 600 million, or almost 1 in 10 people in the world, fall ill after consuming food and water contaminated with viable pathogenic bacterial cells (or spores in the case of botulism) or food containing toxins produced by toxigenic bacteria and moulds (Akter, 2016). Of these, 420 000 people die, including 125 000 children under the age of 5 years. Foodborne illnesses have devastating health implications, such as kidney and liver failure, brain and neural disorders, reactive arthritis, and cancer, with fatal results (WHO, 1996). Although acute gastrointestinal diseases are not all foodborne and foodborne illness does not always result in acute gastroenteritis, food does represent an important vehicle for pathogens causing acute gastroenteritis (Lamin, 2017). Previous research done to assess the quality of street food in several countries has demonstrated that street food was the positive cause of foodborne illnesses (WHO, 2010).

The statistics available in South Africa for the period 2001–2006 show that most epidemics were reported in the provinces of Eastern Cape, KwaZulu-Natal and Limpopo (DoH 2006). According to Shonhiwa *et al*. (2019), 327 foodborne illness outbreaks were reported from January 2013 to December 2017, causing illness in 11 155 individuals, with 78% hospital visits, 4% hospital admissions and 0.4% deaths. Most of the outbreaks were

reported in the warmer months, from KwaZulu-Natal (43%), Gauteng (19%) and Mpumalanga (12%) provinces. Institutional outbreaks were most common (32%), followed by household outbreaks (27%) and community outbreaks (11%). However, Motarjemi (2013) believes that the majority of foodborne illness could be averted if food handlers were knowledgeable in safe food handling and if customers were informed correctly in their choices of food and food handling. Therefore, the aim of the chapter was to assess the microbial levels on vended foods and food surfaces in Mangaung.

3.3 Materials and methods

3.3.1 Study area selections

Microbial samples were collected through non-probability convenience sampling in the Mangaung metro area in Free State, South Africa. The sampling method was chosen to conveniently select businesses that primarily sold cooked meat. Ten whole samples of cooked beef were randomly collected with dry and sterile polythene bags from different vendor stalls at different taxi ranks in Botshabelo, Thaba Nchu and Bloemfontein Central Business District. Three samples were each collected in Botshabelo and Thaba Nchu , in Bloemfontein four samples were collected. In addition, sterile surface swabs were used to collect samples from the preparation surfaces selected on the vending stalls, namely, food preparation areas and meat preparation areas. The samples were collected between 11:00 and 14:00, which is the usual holding period of the food. Afterwards, the samples were kept on ice during transportation to the laboratory. Then, the samples were placed in a refrigerator and analysed without delay.

3.3.2. Microbial quantification

For microbial quantification from meat samples, the beef samples were blended using a sterile blender, and then 10 g of beef samples were mixed with 90 mL of sterile peptone water and homogenised to make a 1:10 dilution. The surface swabs were also mixed with 90 mL of sterile peptone water and homogenised appropriately. Next, serial dilutions were done, the dilutions were prepared up to 10^{-6} dilution, using a spiral plater. A spiral plater is an instrument used to dispense a liquid sample onto the Petri dish in a spiral pattern. The purpose of this method is to inoculate several dilution factors onto a single Petri dish. This allows a significant reduction in the number of dilutions performed and dishes used. About 1 mL of all dilutions was plated in triplicate Petri dishes using plate count agar (PCA). The plates were then incubated at 37 °C for 24 h. In addition, selective media were also used. Baird Parker was used for isolation of *Staphylococcus* species, mannitol egg yolk polymyxin (MYP) agar was used for isolation of *Bacillus* species and xylose lysine deoxycholate (XLD) agar was used for isolation of *Salmonella* species. After the appropriate incubation period, the plates were then examined for microbial growth (Sanders, 2012).

3.3.3. Microbial identification

The species of the foodborne pathogens were identified using a RapID identification kit (Thermo Fisher Scientific, SA). Briefly, organisms were streaked onto selective media Petri dishes for isolation of pure cultures and used the next day. Afterwards, colonies were scraped from the agar plates using an inoculation loop into the RapID inoculation

fluid to achieve the visual turbidity of a 2 McFarland standard. Next, the suspension was mixed thoroughly using the vortex. The contents of the suspension were then transferred to the RapID panel using a pipette. The test suspension was later mixed with the reaction cavities found in the RapID panel. Afterwards, the inoculated panels were incubated at between 35°C to 37°C for 4 h. Following the incubation, two drops of RapID ONE reagent were added to cavities 15, 16 and 17. In additional, two drops of RapID spot indole reagent were added to Cavity 18. The cavities were then observed for colour development. The results of the colour development were recorded using the colour guide sheet and report form provided with the RapID kit. The results were recorded by scoring the colour development using either a plus or minus sign to indicate positive or negative reactions. The observed colour changes were a result of biochemical reactions. The numerical microbe code was then derived from the scores. These microbe codes were entered in the ERIC software for identification of the organism (Thermo Scientific SA).

3.4 Results and discussion

Based on the results found in the current study, Figure 3.1 represents bacterial counts from meat samples collected from the Mangaung Metro street vendors. The microbial levels of the meat samples grown on PCA media are shown as dilution from 1.0 $x10^{-4}$ to 1.1 $x10^{-6}$ CFU/g. Bacterial counts were obtained in Thaba Nchu (≤50 x 10^5 CFU/g), Bloemfontein (≤48 x 10^5 CFU/g), and Botshabelo (≤33 x 10^5 CFU/g). These results are higher when compared to the regulations governing microbiological standards for

foodstuffs and related matters in South Africa, which stipulate that no person should sell meat for which the total colony count of organisms exceeds 10 000 per gram.

These high microbial counts are concerning, as they are an indication that proper food handling and preparation techniques are not always practised by food handlers (Pietrangelo, 2015). Common causes of food poisoning include failing to wash hands completely before preparing or eating food; using utensils, cutting boards or serving dishes that are not clean, causing cross-contamination; consuming dairy products or food containing mayonnaise that have been left out too long; and consuming foods that have not been stored or cooked at the right temperature, especially meat and poultry (Pietrangelo, 2015). Infections caused by microorganisms are largely the result of the poor hygiene of the person responsible for preparing the food (Uçar *et al*., 2016). In addition, lack of good hygiene practices by food handlers has been reported to influence microbial rates and could also place consumers at risk of contracting foodborne illnesses, which may increase the statistical rates of deaths (Setlhare, 2013).

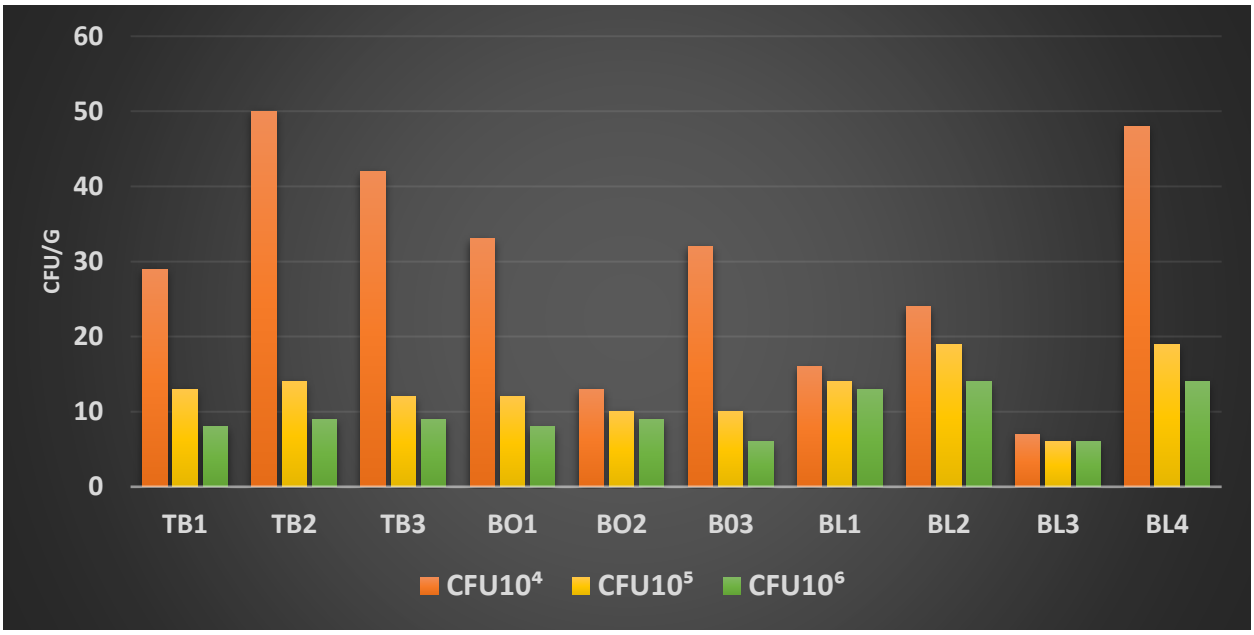

Figure 3.1**:** Total viable counts isolated from meat samples.

Key: TB (Thaba Nchu), BO (Botshabelo), BL (Bloemfontein)

Studies have demonstrated that food safety within the street vending sector is not only affected by food handler practices, the environment in which food is prepared can have a negative impact on food safety and quality. Okojie *et al*. (2014) reported that the preparation of street foods in very dirty surroundings with wastewater and garbage disposed nearby provides nutrients and breeding grounds for rodents and other pests that could cause contamination of the food. Similar results were found in the current study. It was observed from our results that the majority of these vending sites lacked basic infrastructure and services such as potable running water and waste disposal facilities. In addition, hand and dishwashing water was usually insufficient and often reused. In most cases during the survey running water was not available at vending sites; washing of hands and crockery was done in bowls or buckets and sometimes without soap. Furthermore, access to a frequent water supply was limited to facilities that had tap water. All these were considered violations of the regulations governing the general hygiene requirements for food premises and the transport of food and highlight a need for continuous monitoring of these businesses by environmental health professionals.

The regulations governing the general hygiene requirements for food premises and the transport of food (DOH 2002) stipulate that wash basins should always be provided, together with an adequate supply of soap and disposable paper towels. Therefore, street food vending stalls should have potable water available around the stall to make it easy for the vendors to access water. Note that most stalls are erected by vendors, with some erected by government in certain areas; this recommendation could work in areas where

stalls have been erected by government or where stalls are erected close to business areas such as taxi ranks. The regulations also stipulate that no person should handle food intended for consumption if the hands of this person are not washed with soap and water. The lack of basic infrastructure to support the practice of hand washing was a matter of concern. This highlights a possible lack of compliance with proper hygiene practice and implies that the implementation of basic hygiene practices may be difficult because of the lack of hygiene infrastructure.

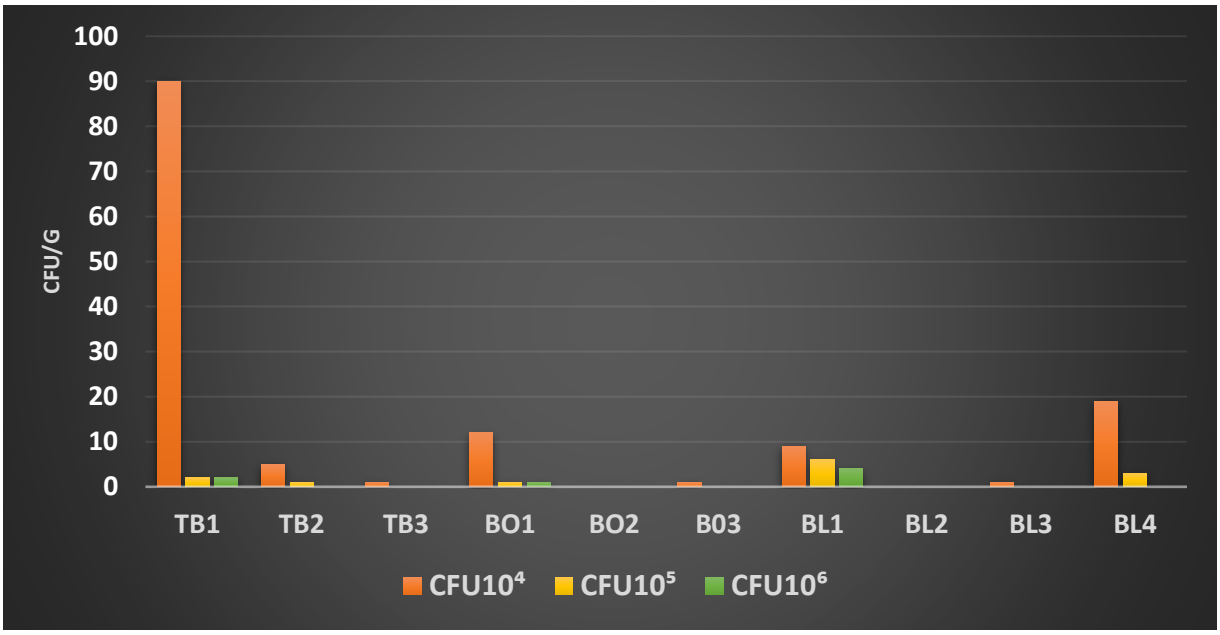

Figure 3.2: Total viable counts from meat samples for *Staphylococcus* isolates.

Key: TB (Thaba Nchu), BO (Botshabelo), BL (Bloemfontein)]

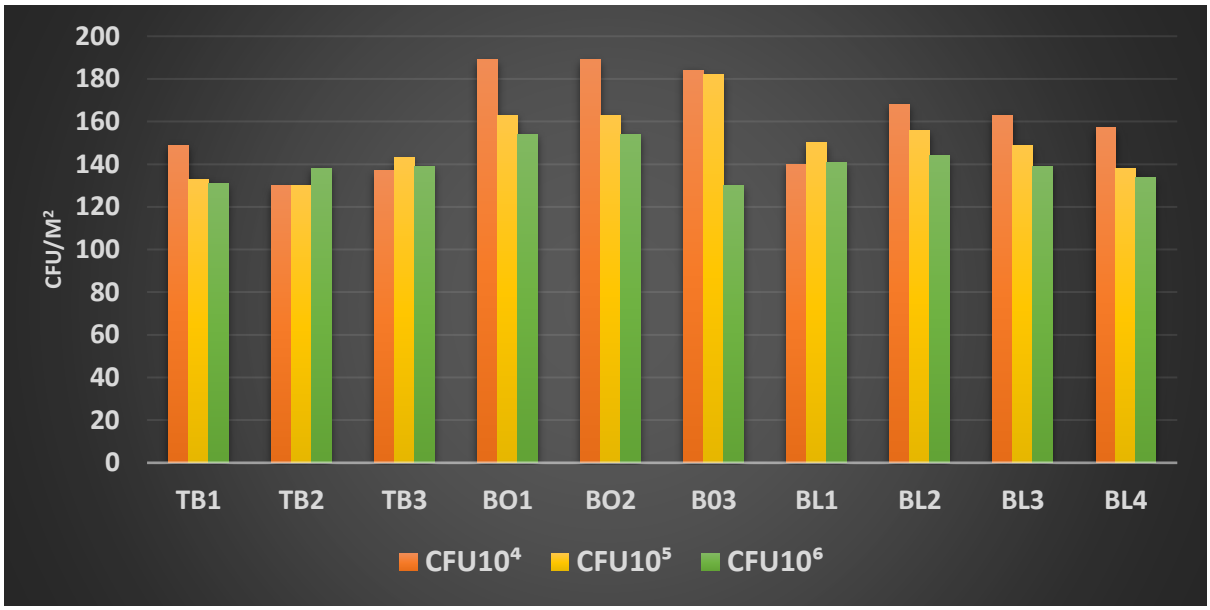

Figure 3.3: Total viable counts from preparation surfaces using microbial swabs.

Key: TB (Thaba Nchu), BO (Botshabelo), BL (Bloemfontein)

Microbial counts from meat samples at Bloemfontein, Thaba Nchu and Botshabelo (Figure 3.1) were found to be higher when compared to the regulations governing microbiological standards for foodstuffs and related matters in South Africa, which stipulate that no person should sell meat for which the total colony count of organisms exceeds 10 000 per gram. The results also showed the presence of *Staphylococcus* isolates (Figure 3.2), indicating that improper food handling practices, handling food with bare hands, reusing surfaces without cleaning first and not wearing aprons contributed to the presence of these foodborne pathogens. *S. aureus* is transferred to food by the person handling it. Persons with skin, nose or throat infections or inflammatory wounds pass this microorganism onto the food (Uçar et al. 2016). The foods posing a particular risk for contamination with *Staphylococcus* spp. include cooked meat, potato salad, desserts with milk, such as custard, and chicken, fish and other meat salads (Duyff 2012). It is important to realise that food vendors can be carriers of pathogens such as *E. coli, S. aureus* and *Salmonella, Shigella* and *Campylobacter* spp. The food vendors eventually transfer these foodborne microbes to the consumers (Siddiqua 2016). Transfer can be direct, from person to person; indirectly in two stages, from person to contact surface and from contact surface to person; and indirectly from person to food and from food to person (Todd et al. 2007). Moreover, a lack of adequate infrastructure can affect the implementation of good hygiene practices.

Figure 3.3 illustrates the total viable counts recorded on food preparation surfaces at various vending stalls in Thaba Nchu, Bloemfontein and Botshabelo. The samples

obtained in Botshabelo (1.1×10^4 CFU/ m^2 – 1.1×10^6 CFU/ m^2) showed a higher microbial count compared to those of Bloemfontein (1.1×10^4 – 1.1×10^5 CFU/ m^2) and Thaba Nchu (1.1×10^4 CFU/ m^2 – 1.1×10^5 CFU/ m^2). This may be attributed to the fact that the vending stalls in Botshabelo are not next to paved roads, as opposed to those in Thaba Nchu and Bloemfontein. The vending stalls were not covered, and there was also dust from construction vehicles working not far from the stalls in Botshabelo. The counts were also found to be higher than the South African national standard (100 CFU/ m^2).

After assessing microbial levels, the RapID identification method was used to identify foodborne pathogens that might be present in meat samples at the selected areas of Mangaung, Metro. The RapID results obtained in Table 3.1 indicate the presence of *Candida guilliermondii* which is yeast and bacteria *Corynebacterium jeikeium, Psychrobacter phenylpyruvicus* and *Peptostreptococcus tetradius,* among others. All these bacteria are found on human skin and might have been transferred from the hands of food handlers to the meat since all the food handlers were observed not to be wearing any gloves during the food preparation and serving (Tables 2.2 and 2.3). Such a food handler may contaminate his or her hands when using the toilet and not washing hands, or bacteria might have spread from raw meat to green salads by the hands of the food handler (Kariuki, 2018).

Staphylococcus aureus was also identified on the meat samples of the three selected areas (Thaba Nchu, Botshabelo and Bloemfontein). The presence of *S. aureus* found in

meat samples highlights the need for providing food handlers with educational training in respect to proper hygiene practices since humans are possible sources of the bacteria. Improper food hygiene practices are worrying as *S. aureus* causes food poisoning with changes in blood pressure and pulse rate (Setlhare, 2013). Staphylococcal disease results from eating food contaminated with toxins such as enterotoxins producing strains of *S. aureus*, which leads to diarrhoea and vomiting. Staphylococci grow in food in which they produce their toxins (DOH, 2011).

Table 3.1: Characterisation of foodborne pathogens isolated from meat samples

Origin Meat sample	Species identified using RapID	Source	Health effects	Reference
Thaba Nchu 1	*Prevotella bivia*	Vaginal tract, oral flora	Endometritis and pelvic inflammatory disease	Mirza *et al*., 2012
Thaba Nchu 1	*Escherichia coli*	Undercooked or raw food, unpasteurised milk, apple juice, contaminated water	Bloodstream infection and urinary tract	CDC, 2011
Thaba Nchu 1	*Psychrobacter phenylpyruvicus*	Human skin	Surgical wound infection	Deschaght *et al.,* 2012
Thaba Nchu 2	*Corynebacterium jeikeium*	Human skin	Infection after disruption of the skin by surgery (cardiac or orthopaedic surgery	Bechara *et al*., 2011
Botshabelo 1	*Escherichia hermannii*	Chickens and humans (blood, urine)	Bloody diarrhoea	Sedlock *et al.,* 2018
Botshabelo 1	*Yersinia kristensenii*	Soil, fresh water, foods,	Human enteritis	Bottone et al., 1977
Botshabelo 2 **Botshabelo 3**	*Peptostreptococcus tetradius*	Human skin	Clinical infections	Song *et al*., 2003

Bloemfontein 1 **Bloemfontein 2**	*Candida guilliermondii*	Human skin and mucosal surfaces	Chronic onychomycosis, septic arthritis and endocarditis	Girmenia *et al.,* 2006
Thaba Nchu 3	*Staphylococcus aureus*	Exterior of a human ear and animals	Human skin infections, sialadenitis and food poisoning	Madigan *et al.*, 2005
Thaba Nchu 2	*Shigella spp*	Salads(potato, tuna, and chicken), raw vegetables, milk and dairy products, and poultry	Blood or mucus in their stool, and they may run a fever	Ghosh *et al*., 2007

Thus, staphylococcal food poisoning does not result from ingesting the bacteria but rather from ingesting the toxins that are already present in the contaminated food (DOH, 2011). Typical contaminated foods include custard, cream-filled pastry, processed meat and fish.

The results obtained (Table 3.1) also indicate the presence of *Escherichia coli* in meat samples isolated from Thaba Nchu. No selective media was used for *E. coli* ,hence it was only picked up from RapID results. The presence of *E. coli* is a cause for concern because the presence of *E. coli* in food usually indicates recent faecal contamination (Kariuki, 2018). Faecal coliforms appear in great quantities in the intestines and faeces of people and animals, hence their presence in a food sample often indicates recent faecal contamination, indicating that there is a greater risk that pathogens may be present (Kariuki, 2018). *Escherichia coli* has also been associated with contaminated water (CDC, 2011). In addition, the water used by street vendors (as observed in Chapter 2) has not been filtered; that is why it may contain bacteria and microorganisms such as *E. coli* species. Moreover, there might have been the presence of *E. coli* in the contaminated water used by the food handlers because they reuse water repeatedly, even when it is no longer sufficiently clean to use (Table 2.6).

In addition to *E.coli*, *Shigella spp*. were isolated from the meat from both Thaba Nchu and Botshabelo (Table 3.1). According to literature, *Shigella* is frequently found in water polluted with human faeces (Dilbaghi *et al*., 2007). They can easily multiply at temperatures between 10 °C and 48 °C (Baş *et al.,* 2007). The optimum growth

temperature for this bacterium is 37 °C (Warren *et al.,* 2007) . Contamination of these foods is usually through the faecal-oral route. Faecal contaminated water and unsanitary handling by food handlers are the most common causes of contamination (Dilbaghi *et al*., 2007). In addition, improper waste disposal has been associated with the transmission of enteric pathogens such as *Salmonella, Shigella* and *E. coli* (Rane, 2011). As demonstrated in Chapter 2, vendors did not have any waste bins near the stalls. Instead, they used boxes and plastic bags to collect waste and placed these near the vending stalls. The presence of *Shigella* might have been from the waste that was observed around the stalls. Hand washing before handling food and thoroughly cooking all food before eating decrease the risk of getting shigellosis (Ram *et al*., 2008).

Prevotella bivia which is found in the vaginal tract was also isolated in the meat sample from the Thaba Nchu selected area. This finding is an indication that the food handlers do not wash their hands after using the toilet. These findings also correspond with observations of food handlers revealed in chapter 2. They all (100%) indicated that the toilets are very far from their stalls and that they have to pay every time they use the toilets (Table 2.4). These findings are consistent with the study conducted in Dhaka by Siddiqua (2016), which indicated that toilets were not available nearby in several cases, thus forcing the vendors to eliminate their body wastes in nearby areas and return to their vending sites without washing their hands.

Based on these results, the Mangaung Metropolitan Municipality are urged to intervene and assist food handlers who currently lack basic infrastructure. This will be one of the key measures to prevent cross-contamination and foodborne pathogens that might lead to foodborne illnesses in the Mangaung Metro.

3.5 Conclusion

Bacterial counts obtained in Thaba Nchu (50 x 10^5 CFU/g), Bloemfontein (48 x 10^5 CFU/g), and Botshabelo (33 x 10^5 CFU/g) showed high microbial counts. These results are alarming when compared to the regulations governing microbiological standards for foodstuffs and related matters in South Africa, which stipulate that no person should sell meat for which the total colony count of organisms exceeds 10 000 per gram.

The results also showed the growth of *S.aureus*, indicating that the improper food handling practices carried out by food handlers contribute to the presence of these foodborne pathogens. The presence of pathogenic organisms such as *E. coli* is a cause for concern as it usually indicates recent faecal contamination. Therefore, consumption of such contaminated street vended foods poses a serious problem to community health.

In the current study, Table 3.1 shows that most of the bacteria identified are usually from the human skin and hair; the results may be an indication that the vendors are not practising food safety measures and hygiene, as transfer from the skin and hair can occur through touching surfaces and lack of hand washing when handling food. Consequently, vendors need to undergo food safety training to limit the spread of microbes through

touching. Moreover, the lack of basic infrastructure (including nearby toilets) and services, such as potable running water and waste disposal facilities, as well as hand and dish washing water that is usually insufficient and often reused by food handlers, play an important role in the presence of foodborne pathogens identified in this study. The surface swabs counts were found to be higher than the South African national standard (100 cfu/m^2), this is a cause for concern because they indicate that the food preparation surfaces are not being properly cleaned by food handlers.

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CHAPTER 4

ANTIMICROBIAL RESISTANCE OF FOODBORNE PATHOGENS INDENTIFIED ON STREET-VENDED FOODS

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4.1 Abstract

Background: The global crisis of antibiotic resistance has reportedly reached a point where, if action is not taken, human medicine will enter a post-antibiotic world and simple injuries could once again be life threatening. Food contamination associated with antibiotic-resistant bacteria is a worrying factor to public health. Antibiotic resistance occurs because bacteria evolve to protect themselves from antibiotics. Recently a large amount of antibiotic-resistant commensal bacteria was detected in many food items, suggesting that the food chain may serve as an important avenue of disseminating antibiotic-resistance to humans.

Materials and methods: Antimicrobial susceptibility testing was done by using the Kirk Bauer disk diffusion susceptibility method. Six antibiotics which included ampicillin (10µg), gentamycin (10µg), ciprofloxacin (5µg), tetracycline (30µg), chloramphenicol (30µg) and streptomycin (10µg) were used to determine antimicrobial susceptibility.

Results: About 80% isolates of fourteen organisms showed sensitivity to ciprofloxacin, streptomycin, tetracycline and gentamycin. However, *Staphylococcus aureus* isolates showed resistance to gentamycin (100%), streptomycin (63%) and tetracycline (68%), while *Escherichia hermannii* showed 50% resistance to ampicillin followed by *Escherichia coli* which showed 100% resistance to ampicillin.

Conclusion: The presence of antibiotic-resistant isolates detected in this study indicates that street foods in the Mangaung Metro might pose a problem to public health. These foods could expose consumers to antibiotic-resistant bacteria and over a long period of exposure this could become a public health risk. Therefore, collaboration with Mangaung

environmental health practitioners is needed to train food vendors on how to produce safe foods that will reduce the incidence of foodborne diseases and the transfer of antibiotic-resistant microorganisms from food to man. The ideal would be to raise the awareness of unwholesome practices in street food trading through audience participatory programmes. Moreover, a law should be enforced that states that every street vendor has to undergo training and be warded a certificate before he or she may start up a vending business.

Keywords: Antimicrobial resistance, foodborne pathogens, street food

4.2 Introduction

Street-food vending and fast-food enterprises are two of the major businesses that contribute to the socio-economic development in many countries (Rane, 2011; Ogidi *et al*., 2016). Street foods and fast foods are the preferred choices of many people, especially urban dwellers, because the food is ready-to-eat, cheap, convenient and accessible as an immediate want (Rane, 2011; Ogidi *et al.*, 2016). Most of the ingredients used in fast food are nutritious. However, they may harbour various pathogenic organisms, including *Escherichia coli, Enterobacter* spp*. Staphylococcus aureus, Salmonella* spp. and *Listeria monocytogens* (Ahmed *et al*., 2014).

Street foods play a significant role in transferring foodborne illnesses to consumers (Sabuj *et al*., 2018). Food contamination with antibiotic-resistant bacteria can also be a major threat to public health, since the antibiotic resistance determinants can be transferred to other pathogenic bacteria potentially comprising the treatment of severe bacterial infections (Eromo *et al*., 2016). Current reports indicate that most of bacteria are resistant to antibiotics and it is very difficult to control them, sometimes reaching such levels that can cause the death of an individual (Sabuj *et al*., 2018).

Antibiotic resistance (AR), which is defined as the ability of an organism to resist the killing effects of an antibiotic to which it was normally susceptible, has become an issue of global interest (Agyare *et al*., 2018). The use of antimicrobial agents in animals and more

importantly, in food-producing animals has important consequences for both human and animal health as it can lead to the development of resistant bacteria (Ayukekbong *et al*., 2017). These resistant bacteria (with resistance genes) in animals can be transferred to humans through the consumption of food, through direct contact with food-producing animals or through environmental spread (Ayukekbong *et al*., 2017).

Effort has been directed to study the antibiotic resistance of the non-pathogenic bacteria, most often *E. coli* (Kirbis *et al*., 2015). *E. coli* is a normal inhabitant of the warm-blooded animal intestine, including the human intestine and can easily contaminate food products during animal evisceration at slaughter or during food manipulation (Wang *et al*., 2012). The recent discovery of the large pool of antibiotic-resistant bacteria in many retail food items suggests that the general public is constantly exposed to antibiotic-resistance through daily food consumption. Moreover, the food chain may serve as a crucial avenue connecting the antibiotic-resistance between the environment and human beings (Wang *et al*., 2012). This study aims to investigate the prevalence of antibiotic resistance in bacteria isolated from street-vended food of the Mangaung Metro.

4.3 Materials and methods

4.3.1 Antimicrobial susceptibility testing

The Kirby-Bauer disc diffusion method described by Bauer *et al*. (1966) was used to perform an antimicrobial susceptibility test on Mueller Hinton agar using the following antibiotic disks: ampicillin (10 µg), gentamycin (10 µg), ciprofloxacin (5 µg), tetracycline 30 µg), chloramphenicol (30 µg) and Streptomycin (10 µg). The inoculum was prepared from 18 hour-old broth culture of each isolate and isolate absorbance was adjusted to 0.5 McFarland equivalent. The inoculum was spread over a dried surface of a Milton Huller agar plate by streaking with a swab three times over the entire agar surface. The plate was rotated at approximately 60 degrees each time to ensure an even distribution of the inoculum. Afterwards, the plates were left to dry for five minutes. The antimicrobial-impregnated disks were then placed on the surface of the agar, using forceps to dispense each antimicrobial disk one at a time. The forceps were sterilised by cleaning them with a sterile alcohol pad and allowing them to air dry. Then, one disk was removed from the cartridge using the forceps. The disk was placed on the agar plate by gently pressing the disk with the forceps to ensure complete contact with the agar surface. Six different disks were placed on each agar plate. Once all the disks had been placed on the agar plates, the plates were incubated at 35 °C for 18 hours.

4.3.2. Data analysis

After the antimicrobial agent had diffused into the agar and inhibited germination and the growth of the tested microorganism, diameters of inhibition growth zones were measured. The inhibition areas on the agar plates were measured using a spiral platter. The results

obtained by the spiral platter were compared to the Clinical and Laboratory Standard Institute (CLSI) guidelines.

4.4 Results and discussion

The diameters of inhibition growth zones were measured. A zone of inhibition is defined as clear region around the paper disc saturated with an antimicrobial agent on the agar surface. Isolates were classified as susceptible, intermediate or resistant according to the zone diameter interpretative standards recommendations from the Clinical and Laboratory Standards Institute (CLSI, 2005) and recorded as susceptible, intermediate, or resistant to each antibiotic tested. The susceptible (S) category implies that isolates are inhibited by the usually achievable concentrations of antimicrobial agent when the recommended dosage is used for the site of infection. The intermediate (I) category implies clinical efficacy in body sites where the drugs are physiologically concentrated (e.g. quinolones and beta-lactams in urine) or when a higher than normal dosage of a drug can be used. The resistant (R) category implies that isolates are not inhibited by the usually achievable concentrations of the agent with normal dosage schedules, or that demonstrate zone diameters that fall in the range where specific microbial resistance mechanisms (e.g. beta-lactamases) are likely, and clinical efficacy of the agent against the isolate has not been reliably shown in treatment studies (CLSI, 2005).

Percentage results for this study were determined by dividing the measured inhibition zone (mm) from agar plates with zone diameter interpretative standards recommendations from the Clinical and Laboratory Standards Institute (CLSI, 2005) on Table 4.1. Based on the results found, the antimicrobial susceptibility test results (Table 4.2) revealed that out of all the fourteen organisms, 80% of them (*Candida guilliermondii, Prevotella bivia, Psychrobacter phenylpyruvicus, Yersinia kristensenii ,Corynebacterium jeikeium and Pepetostrptococcus tetradius)* showed sensitivity to ciprofloxacin, streptomycin, tetracycline and gentamicin. The results indicate that these drugs can be used as drugs of choice when treating foodborne illness.

Current results indicated that chloramphenicol showed intermediate zones of inhibition on seven of the fourteen isolates (Table 4.2). ampicillin showed intermediate zones of inhibition on only four of the fourteen isolates (Table 4.2). These intermediate zones of inhibition means that a higher dose of these antibiotics is needed to prevent growth of the bacteria or to treat foodborne illness caused by these bacteria. *S. aureus* isolates showed resistance to gentamycin (100%), streptomycin (63%) and tetracycline (68%) which would make the treatment of foodborne infections difficult. We belive this is a first report that demonstrates susceptibility or resistance of bacterial isolates from food environments in the Mangaung region.

Table 4.1: Zone diameter interpretative standards recommendations from the Clinical and Laboratory Standards Institute (CLSI, 2005). (Note that there are different charts for different organisms)

	Resistant	Intermediate	Susceptible
Cefazolin (30 µg)	≤14	15-17	≥18
Clindamycin (2 µg)	≤14	15-20	≥21
Erythromycin (15 µg)	≤13	14-22	≥23
Gentamicin (10 µg)	≤12	13-14	≥15
Oxacillin (1 µg)	≤10	11-12	≥13
Penicillin G (10 µg)	≤28	--	≥29
Tobramycin (10 µg)	≤12	13-14	≥15
Vancomycin (30 µg)	--	--	≥15

Table 4.2: Antimicrobial susceptibility results obtained by the spiral plater.

Origin of Meat Sample	Organism	Antibiotics					
		AMP	GEN	TET	CIP	STR	CHL
Thaba Nchu 1	*Prevotella bivia*	I	S	S	S	S	I
Bloemfontein 1 **Bloemfontein 2**	*Candida guilliermondii*	S	S	S	S	S	I
Thaba Nchu 1	*Escherichia coli*	R	S	S	S	I	I
Botshabelo 2 **Botshabelo 3**	*Peptostreptococcus tetradius*	I	S	S	S	S	I
Botshabelo 1	*Escherichia hermannii*	R	S	I	I	S	S
Thaba Nchu 2	*Corynebacterium jeikeium*	S	S	S	S	S	S
Botshabelo 1	*Yersinia kristensenii*	S	S	S	S	S	S
Thaba Nchu 1	*Psychrobacter phenylpyruvicus*	S	S	S	S	S	S
Thaba Nchu 2	*Shigella* spp. *1*	R	S	I	S	S	I

Botshabelo 2	*Shigella* spp. *2*	S	S	S	S	S	I
Thaba Nchu 3	*Staphylococcus aureus 1*	I	S	S	S	S	S
Botshabelo 1	*Staphylococcus aureus 2*	I	S	S	S	S	I
Bloemfontein 1	*Staphylococcus aureus 3*	S	R	S	S	R	S
Bloemfontein 4	*Staphylococcus aureus 4*	S	S	R	S	R	S

In another study, Loeto *et al*. (2017) reported that the highest resistance rate by *S. aureus* was observed for tetracycline (38.1%), followed by streptomycin (26.2%). Islam *et al*. (2019) also reported that *S. aureus* was resistant to tetracycline (11%) and 3% to gentamycin. Streptomycin and gentamycin belong to a class of antibiotics called aminoglycosides. Aminoglycosides inhibit the synthesis of proteins in bacteria, eventually leading to cell death. They are only effective against certain Gram-negative bacteria, as well as some Gram-positive bacteria, but are not absorbed during digestion, so must be injected. In the present study, *S. aureus* was found to be resistant to two antibiotics from aminoglycosides, this finding means that *S. aureus* might be resistant to all the antibiotics that belong to the aminoglycosides class. A study conducted by Mahdiyoun *et al*. (2016) in Iran also indicated a high resistance rate of MRSA against aminoglycosides. Aminoglycosides are one of the classes of antibiotics that play an important role in the treatment of staphylococcal infections (Khosravi *et al*., 2017). These are often used synergistically in combination with either beta-lactam or glycopeptides, especially for the treatment of complicated staphylococcal infections (Khosravi *et al*., 2017). However, today MRSA has acquired multiple resistance to a wide range of antibiotics, including aminoglycosides (Khosravi *et al*., 2017).

In addition, 62% of *Shigella* spp. showed resistance to ampicillin, followed by *Escherichia hermannii* which showed 50% resistance to ampicillin and lastly, *E. coli* which showed 100% resistance to ampicillin. This indicates that ampicillin will no longer be used for treatment of any infection caused by *Shigella, E. hermannii* and *E. coli* isolated in the

current study. A marked resistance of microorganisms to commonly used antibiotics such as tetracycline, gentamicin, chloramphenicol, ciprofloxacin, streptomycin was reported to be associated with the coexistence of resistance genes with mobile elements, such as plasmids, transposons and integrons (Thong *et al*., 2011; Ogidi *et al*., 2016).

The transfer of antibiotic-resistant genes in food products and the environment could be directly or indirectly linked to human activities, such as the use of antibiotics in farming to produce some edible foods (Economou *et al.,* 2015). The presence of antibiotic resistant *S. aureus* in the present study indicates improper food handling practices were somehow carried out by food handlers to contribute to the presence of these foodborne pathogens. These non-compliance practices may contribute to the spread of antibiotic-resistant bacteria. Moreover, *Corynebacterium jeikeium, Psychrobacter phenylpyruvicus* and *Peptostreptococcus tetradius,* amongst others, were present in street food. All these bacteria are found on human skin and might have been transferred from the hands of food handlers to the meat since all the food handlers were seen not to be wearing any gloves during the food preparation.

Antibiotic-resistant *Shigella* was also present in the meat. This means that some of the food handlers may have had Shigellosis while handling food. Shigellosis is defined as acute diarrhoea that may progress to bloody mucoid diarrhoea (Ranjbar *et al*., 2019). Conventional antibiotic therapeutics against shigellosis have become increasingly inefficient owing to the increase in the number of MDR strains (Ranjbar *et al*., 2019). The

emergence of antibiotic-resistant microorganisms from foods raises concern as most of the resistant strain has spread into other environments where they can infect man through some conscious or unconscious activities (Ogidi *et al.*, 2016) because much of our food is derived from farmed animals, a feature that links human and animals alike. The food chain and the broader environment provide a convenient route by means of which humans can become infected with AMR bacteria. Therefore, from the results obtained in this study, food vending could also be another activity that plays a role in spreading antibiotic-resistant genes in the environment. The focus of future studies could involve sampling water from that particular environment and checking whether similar antibiotic-resistant genes are to be found. In addition, future investigation should be done on the environment where the animals are being kept comparing the results with those of the present study. This would then be followed by investigating mechanisms of resistant genes using genome sequencing.

4.5 Conclusion

The presence of antibiotic-resistant isolates detected in this study provides some evidence that street foods might pose a major problem for public health. *Staphylococcus aureus* isolates showed resistance to gentamycin (100%), streptomycin (63%) and tetracycline (68%). *S. aureus* has garnered substantial public health attention owing to its multi-drug resistance which accounts for increased mortalities in health facilities (Waters *et al*., 2011). *Escherichia hermannii* showed 50% resistance to ampicillin, followed by *Escherichia coli* which showed 100% resistance to ampicillin. Results from this study

indicate that the general public can easily be exposed to antibiotic-resistant bacteria daily through conventional food intake because the street vendors are non-complaint with food safety hygiene practices. Exposure of consumers to antibiotic-resistant bacteria may make them vulnerable to various food diseases caused by antibiotic-resistant pathogens. Some of these diseases are fatal. The WHO (2014) estimated that infections accounted for 45% of deaths in Africa and South-East Asia and that these diseases were responsible for 48% of premature deaths worldwide. It is therefore important that the street vendors are given proper food safety training. Collaborative work with environmental health practitioners needs to introduce good manufacturing practices (GMP) training to the food vendors in order to produce safe and wholesome foods. This training should be conducted in the local language to ensure that the vendors understand exactly what it is expected of them. The ideal would be to raise awareness of the threat of unwholesome practices in street food trading through audience participatory programmes. Professionals in food and health-related disciplines should be engaged to draw up guidelines for the management of street food practices' implementation of the hazard analysis and critical control points (HACCPs) concept along the entire chain of the business, educating vendors and consumers alike on hygiene and safe food practices. Additionally, a law should be enforced that stipulates that every street vendor should undergo training and be awarded a certificate before he or she starts up a vending business.

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CHAPTER 5

GENERAL DISCUSSION, CONCLUSIONS AND RECOMMENDATIONS

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5.1 Introduction

In general, there is scientific evidence demonstrating inadequate hygiene and poor quality of street foods in the African region (Kariuki, 2018). Microbial agents such as *E. coli*, *S. aureus*, and *Salmonella* have caused foodborne disease epidemics, thereby highlighting problems with the safety of food (Kariuki, 2018). This has also raised public apprehension that food processing and modern farming systems do not provide adequate safety measures to ensure public health. Factors which contribute to potential hazards in foods include poor hygiene, improper agricultural practices at all stages of the food chain, lack of preventive controls in food processing and preparation operations, contaminated raw materials and insufficient or inappropriate storage, among others (Mutangadura, 2004). Foodborne illnesses have been previously reported as a major cause of death in developing countries and a significant international health problem associated with food safety (Mutangadura, 2004). Additionally, among other cases, episodes of diarrhoea related to the consumption of ready-to-eat street foods have been reported (Eromo *et al*., 2016). The prevalence of antimicrobial resistance among foodborne pathogens has increased during recent decades (Eromo *et al*., 2016), adding to the existing challenges within the food safety sector. For example, a study in Benin revealed that 15.18% of *S. aureus* stains isolated from street foods were resistant to methicillin (Sina *et al*., 2011). Another study conducted by Sabuj *et al*. (2018) in Bangladesh reported that from antimicrobial susceptibility test results, 70-100% of *S. aureus, Salmonella and E. coli* showed resistance to ampicillin, amoxicillin, nalaxidin acid, tetracycline and erythromycin which would make treatment of infections difficult. In Ethopia, Amare *et al*. (2019) reported that *Citrobacter* species showed 100% sensitivity to gentamicin, ceftriaxone, and

tetracycline. In addition, *S. aureus* showed the highest resistance against penicillin (73.53%). Moreover, *E. coli, Enterobacter and Citrobacter* species are resistant to ampicillin (86.67, 70 and 75%, respectively). The presence of antibiotic-resistant bacteria in street-foods has become a global health concern (Dey et al., 2018).

5.2. General discussion

During observations as discussed in Chapter 2, food vendors' attitude towards food safety was positive, although some of the actual food-handling practices by street vendors raised some serious concerns. The respondents also indicated that they seldom had inspections, discussions, or training from the environmental health practitioners (EHPs) regarding food safety matters. Hence, during observations most of the meat handlers were found to handle food with their bare hands. Another observation showed a lack of storage around the vending stalls. Due to this, vendors are forced to travel with their equipment at the end of the day and in the mornings. Moreover, the vending stalls were found not to be conducive to either vendors or consumers. The stalls were not properly covered; they were exposed to dust, vehicle exhaust fumes and insects.

A further problem that was highlighted during the questionnaire survey was that toilets were very far from their vending stalls. The distance between the stalls and toilets has been associated with foodhandlers' failure to adhere to washing hands thoroughly after using the toilet (Karuiki, 2018). There was also a lack of critical facilities for food safety

(potable water and electric supplies, waste disposal services, drainage) around the stalls. The absence of water points near their workplaces and poor drainage facilities makes it difficult for vendors to safeguard food safety and hygiene. Additionally, the absence of these facilities leads to high pest infestation, ultimately resulting in increased risk of microbial contamination. However, in the current study, no pests were observed around the vending stalls. It must be noted that the study was conducted during the winter season which might be the reason for the absence of flies around the stalls.

After assessing knowledge, attitudes, and practices among street vendors in the Mangaung Metro by means of questionnaires and a checklist (Chapter 2), a determination of the microbial levels of the food samples and surface swabs was undertaken and analysed in the laboratory. Surface swabs results obtained in Chapter 3 indicated the presence of foodborne pathogens. These results are a cause for concern, as they were found to be higher than the national standard (100 CFU/m^2).

Other than food preparation surfaces, microbial counts of meat samples obtained in Bloemfontein, Thaba Nchu and Botshabelo were also found to be lower than the national standard (100 CFU/g) as seen in Chapter 3. Nevertheless, the results showed (Figure 3.2) growth on *S. aureus,* indicating that the improper food handling practices carried out by food handlers contribute to the presence of these foodborne pathogens.

After assessing microbial levels on meat samples, the identification of bacteria was performed using the RapID system. The predominant species identified were *S. aureus* and *E. coli*. The presence of these species is a major cause of concern because *S. aureus* and *E. coli* are the most common bacteria that play a role in human diseases (Setlhare, 2013) and are commonly known as antibiotic-resistant pathogens (Amare *et al*., 2019).

Findings in Chapter 4 revealed that *S. aureus* isolates showed resistance to gentamycin (100%), streptomycin (63%) and tetracycline (68%) which would make the treatment of infections difficult. Moreover, about 62% of *Shigella* spp. showed resistance to ampicillin, followed by *Escherichia hermannii* which showed 50% resistance to ampicillin. Lastly, *Escherichia coli* showed 100% resistance to ampicillin. This indicates that ampicillin may no longer be used for treatment of any infection caused by *Shigella, E. hermannii and E. coli*. Hussen *et al*. (2019) conducted a study in Ethiopia and reported that *Shigella* species were 95% resistant to ampicillin. *Escherichia coli* also showed resistance to ampicillin in a study conducted by Sabuj *et al*. (2018) in Bangladesh.

According to (Kimera et al , 2020) , there are very high levels of antimicrobial use and antimicrobial resistance, especially for tetracycline, aminoglycoside and penicillin in animal production systems in Africa. This is likely to escalate the already high prevalence of antimicrobial resistance and multi drug resistance in the continent. This, coupled with weak regulations and antimicrobial resistance surveillance systems in the region is a great concern to the animals, environment and humans as well (Kimera et al , 2020). It is

therefore important to enforce strict regulation procedures which will also assist in preventing the spread of antibiotic resistant bacteria .The development of standards which are legally enforceable becomes important to provide a policy and legal framework for reducing the risk of the phenomenon spreading in South Africa.

Therefore, from the results obtained in this study, it is recommended that future investigation should be conducted in the environment where the animals are being kept comparing those results with the present study. This would then be followed by investigating mechanisms of resistant genes using genome sequencing.

5.3 Conclusion and recommendations

Despite street vendors' positive attitude towards food safety, instances of non-compliance were observed. Some of the vendors handled food with their bare hands and did not wear aprons during the processing and serving of food. Food safety is a shared responsibility amidst government departments, establishment managers and workers. However, food handlers' knowledge of personal and general hygiene plays a pivotal role in the prevention of cross-contamination. Since most vendors in this study did not have any food safety training (Chapter 2), there is a need to organise formal training in food safety for the street food vendors. It is recommended that EHPs should make use of the Five Keys to Safer Food behavioural methodology as a guide for training on principles of good hygiene practices. Following that, certificates should be issued to food vendors who have

satisfactorily completed their training in basic food hygiene and sanitation. The ideal would be to raise the awareness of the threat of unwholesome practices in street-food trading through audience participatory programmes. The engagement of professionals in food- and health-related disciplines should be considered to draw up guidelines for the management of street food practices' implementation of the hazard analysis and critical control points concept along the entire chain of the business, as well as the education of vendors and consumers on hygiene and safe food practices. Furthermore, a law should be enforced that stipulates that every street vendor must undergo training and be awarded a certificate before he or she starts up a vending business.

From the results obtained in this study, it can be concluded that the lack of adequate infrastructure also plays a significant role in the increase of microbial contamination. There is a need for the government to make more infrastructure available such as potable water, toilets and waste disposal facilities since the lack of these structures was evident in the current study. The lack of facilities highlights the fact that the city authorities either do not plan for the informal food sector or have plans but are not able to implement them. It must be noted that some of the vendors occupy the spaces illegally. Furthermore, street food vendors should be officially recognized as an economic contributor and, where possible, be included in urban development programmes. It is recommended that the city authorities provide the street vendors with food stalls that will protect the food from dust. If potable water is not available, a suitable source of safe water should be used.

One of the identified limitations in this study was that no database appears to have been used previously to capture the number of street vendors operating in the Mangaung Metro. As this information is unavailable, the sample size was affected. Therefore, it is recommended that the Mangaung Metro municipality creates a database that captures the details of all operating street vendors. The EHPs should visit the existing stalls and request the vendors to register their business and give them a certificate of acceptance. This certificate should be displayed on the cart or kiosk or somewhere around the stall for verification. Registering or licensing vendors has many advantages for authorities. It enables authorities to identify persons employed in such enterprises and the types of food sold, to raise revenue and to provide an opportunity to give food handlers training in food safety. It was recognised that policies and regulations for safe street-food trade are very weak and poorly enforced in most developing countries and even non-existent in some countries (Zhang *et al.*, 2014). The findings of the current study are in agreement with the latter statement. The bylaws and legislations are weak and need to be strengthened (Alimi ,2016) . This would undoubtedly ensure a significant reduction in the hazards of street food consumption (Alimi ,2016)

Furthermore, the global crisis of antibiotic resistance related to foodborne pathogens has reached a point where, if action is not taken, human medicine will enter a post-antibiotic world and simple injuries could once again be life threatening (Meek *et al*., 2015). The fate of antibiotic resistant bacteria in remote ecosystems is largely unknown but the emergence of resistance genes in these remote environments has the potential to

decrease antibiotic efficacy in human medicine and agriculture. It has already been noted that wild animals and their surrounding environments have been shown to be important reservoirs of antibiotic resistant bacteria. Furthermore, biodiversity in the natural, more remote environments, could be threatened by ABR pollution (Mercat et al., 2016). It is not just a human health issue. New antimicrobial agents are needed urgently. However, better use of existing agents is equally important. More appropriate use of antibiotics in medicine is vital, but the extensive use of antibiotics outside medical settings is often overlooked.

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Appendix 1: Assessment of vendors' food safety knowledge

Statement	Response: Yes	Response: No
1. It is important to wash your hands before handling food		
2. It is important to wear protective clothes while cooking		
3. Knives and cutting boards should be properly sanitised to prevent cross-contamination		
4. Raw and cooked foods should be stored separately to reduce the risk of food contamination		
5. Do you have storage facilities (refrigerator) for both raw and cooked meat?		
6. Do you know the correct temperature for the refrigerator?		
7. Do you reheat your food before serving?		
8. Food prepared in advance reduces the risk of food contamination		
9. Dish towels can be a source of food contamination.		
10. If you could improve the working conditions here, what would you do?		

_____
_____
_____
_____
_____

11. How would this help improve the safety of your food?

_____
_____
_____

Appendix 2: Assessment of vendors' waste management

Statement	Response: Yes	Response: No
1. Are dust bins available for customers use?		
2. Do you have a drainage system available for disposal of liquid wastes?		
3. Do you have your own waste bin or do you share it with other vendors?		
4. How often does municipality collect waste?		
5. Do you experience problem of waste disposal during strikes by waste collectors?		
6. How far from the vendors' stalls are the toilets located?		

7. What challenges are you facing with regard to waste management?

8. What can be done to improve the environment around your stall?

Appendix 3: Assessment of water availability around the stalls

1. What source of water do you use?

_____
_____
_____

2. Is the source around or close to the vending stall?

_____
_____
_____

3. Is water always accessible?

_____
_____
_____

4. What kind of container is used?

_____
_____
_____

5. If buckets are used, for how long do you store the water?

_____
_____
_____

6. How often do you change the dish-washing water in a day?

_____
_____
_____

7. Where do you dispose of the wastewater?

_____
_____
_____

8. What are you're the challenges that you face on a daily basis as a street vendor?

_____
_____
_____
_____

9. What would you like to see change in the near future?

_____
_____
_____
_____

Appendix 4. Observation list for street vendors' assessment

Table 1. Food-handling practices

Observation	*Yes*	*No*
Food prepared at the stall		
Wash hands with soap before cooking		
Wash food before cooking		
Preparation surface clean		
Prepared on same surfaces more than twice		
Use of apron		
Long nails		
Wears jewellery		
Handles food with bare hands		
The vendor's clothes are clean and presentable		
Hair covering		

Table 2. Water availability around the stalls

Observation	*Yes*	*No*
Source of water at the stall/nearby		
Drainage for disposal of wastewater at the stall/nearby		
Hand washing facilities available nearby		
Water stored in clean containers		
Water reused more than twice		

Table 3. Waste management around the stalls

Observation	*Yes*	*No*
Trash bins available for customer use		
Enough trash bins available for disposal of solid wastes		
The environment around the stall is clean: far from rubbish		
Presence of vectors e.g. cockroaches, rodents, around the stall		
Stalls are located far from toilet facilities		
Vending stall protected from sun, wind and dust		
Location of the business interferes with human or traffic flow		

Appendix 5. Letter to Mangaung Metro Municipality

Central University of Technology, Free State

Malerato Moloi
507 Marhei court
Bloemfontein
9300

Mangaung Metropolitan Municipality
Bram Fischer Building
Nelson Mandela and Markgraff Street
Bloemfontein
9300

Dear Sir / Madam

I am Malerato Moloi, a Masters students from Environmental health department in Central University of Technology. I am aiming to work on a study investigation the prevalence of antibiotic resistant bacteria in street vended foods. The objective of the study also includes collection of food samples from the vendors in order to determine the bacteria levels of food and the facility on which foods is prepared.

I hereby request permission to conduct this study involving the contact with street vendors. Additionally, I request that the municipality allocate one Environmental health practitioner to assist in making contact with the street vendors for corporation.

Yours Sincerely

Malerato Moloi

MANGAUNG METRO MUNICIPALITY
2 9 -11- 2019
MUNICIPAL HEALTH SERVICES
BUSINESS LICENSING OFFICE

Appendix 6. Research consent form

Bloem: Nobengazi; Mbizo

RESEARCH CONSENT FORM

Title: The prevalence of antibiotic resistant bacteria in street vended foods

The objective of this study is to collect the chicken samples from the street vendors to determine if the samples are contaminated by antibiotic resistant bacteria or not. Additionally, the surface on which the foods are being prepared will also be assessed for the presence of bacteria. This research also seeks to determine the factors that contribute to contamination of street vended foods

Participants are being asked to partake in this study because they own or work at a vending stall. Participants must ensure they understand what the research is all about and ask questions whenever they are unsure. Participants have the right to withdraw at any time without providing a reason. The names and personal details of the participants will not be revealed. all the documentation and information obtained during the study will be kept confidential.

By participating on this study and providing us with the food surface samples will assist the understanding and determining the presence of bacteria in street vended foods. The participation will also give us and idea of the challenges faces by the street vendors on daily basis and how to improve those challenges.

This consent form will be translated into a preferred language to the participants. The findings of this study as well as the recommendations for the participants will be communicated to the participants by form of writing. Mangaung metro municipality is aware of this study and the results will also be shared with them. The participants will be given training on how to ensure that their food is microbiologically safe.

Name of the researcher: Malerato Moloi

Position of the researcher: Master's student

Contact details of the Researcher: - Email address maleratomoloi@yahoo.com

- Cell phone number: 0731832525

		Please Initial Box
1.	I confirm that I understand what the research is about and its purpose . I has the opportunity to ask questions and agree to participate	☑
2.	I am aware that my participation is entirely voluntary and that I can withdraw at any time without providing a reason.	☑